Harris, A.
09/508849

-key terms claim 1

```
     FILE 'REGISTRY' ENTERED AT 10:31:36 ON 26 APR 2001
                E PROTEASE/CN 5
L1          261 S PROTEASE ?/CN
                E PROTEINASE/CN 5
L2         2656 S PROTEINASE ?/CN
L3         2864 S L1 OR L2
                E FAS LIGAND/CN
L4            8 S FAS LIGAND ?/CN

     FILE 'CAPLUS' ENTERED AT 10:33:55 ON 26 APR 2001
L5         2989 S FAS(W)(L OR LIGAND) OR FASL OR L4
L6         4189 S (L3 OR PROTEASE OR PROTEINASE)(S)RESIST?
L7           21 S L5 AND L6

L7   ANSWER 1 OF 21  CAPLUS  COPYRIGHT 2001 ACS
ACCESSION NUMBER:         2001:185915  CAPLUS
DOCUMENT NUMBER:          134:231888
TITLE:                    Protease-resistant analogs
                          of Fas ligand inhibitory
                          protein (FLINT) complexed with divalent metal
                          cation and uses therapeutic thereof
INVENTOR(S):              Atkinson, Paul Robert; Tian, Yu; Witcher,
                          Derrick Ryan
PATENT ASSIGNEE(S):       Eli Lilly and Co., USA
SOURCE:                   PCT Int. Appl., 44 pp.
                          CODEN: PIXXD2
DOCUMENT TYPE:            Patent
LANGUAGE:                 English
FAMILY ACC. NUM. COUNT:   1
PATENT INFORMATION:

     PATENT NO.        KIND  DATE           APPLICATION NO.  DATE
     ---------------   ----  --------       ---------------  --------
     WO 2001018202      A2   20010315       WO 2000-US20806  20000831
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH,
             CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH,
             GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK,
             LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ,
             PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ,
             UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU,
             TJ, TM
         RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH,
             CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE,
             BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
PRIORITY APPLN. INFO.:                    US 1999-153433   P  19990910
AB   The invention provides a compn. comprising a protease-
     resistant human Fas ligand inhibitory
     protein (FLINT) analog or fusion protein thereof complexed with a
```

divalent metal cation (such as Zn++, Ni++ or Ca++). The invention addnl. provides parenteral pharmaceutical formulations comprising the FLINT-cation compds. and methods of using such compds. for treating or preventing diseases and disorders, e.g. those that may be assocd. with the binding of Fas to Fa5L, and/or LIGHT to LTPR and/orTR2/HVEM receptors. The invention further provides a process of prepg. such compds., which comprises combining a **protease -resistant** FLINT analog or fusion protein comprising a FLINT analog and a divalent metal cation in an aq. soln. at a pH of about 4.5 to 9.0.

IT **210227-94-4**
RL: PRP (Properties)
(unclaimed protein sequence; **protease-resistant** analogs of **Fas ligand** inhibitory protein (FLINT) complexed with divalent metal cation and uses therapeutic thereof)

L7 ANSWER 2 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2001:87557 CAPLUS
DOCUMENT NUMBER: 134:250117
TITLE: Transition of apoptotic resistant vascular smooth muscle cells to troptotic sensitive state is correlated with downregulation of c-FLIP
AUTHOR(S): Imanishi, Toshio; Hano, Takuzo; Nishio, Ichiro; Liles, W. Conrad; Schwartz, Stephen M.; Han, David K. M.
CORPORATE SOURCE: Division of Cardiology, Department of Medicine, Wakayama Medical College, Wakayama City, 641-8510, Japan
SOURCE: J. Vasc. Res. (2000), 37(6), 523-531
CODEN: JVREE9; ISSN: 1018-1172
PUBLISHER: S. Karger AG
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Fas and its ligand, **FasL**, are a receptor-ligand pair identified as promoting cell death in several tissues. Vascular smooth muscle cells (VSMCs) are resistant to **FasL** or anti-Fas antibody (Ab) signal, and a no. of in vitro studies show that VSMC death can only be induced by anti-Fas Ab or **FasL** in the presence of protein inhibitor or addnl. inflammatory mediators. It remains to be clarified whether known, constitutively expressed cytoprotective mols. are reduced by protein inhibitor, thereby accounting for sensitization to cell death by Fas/ **FasL** signaling. We found that Fas mRNA and protein exist in several primary VSMCs, as previously reported. We also demonstrated (1) that crit. death-signaling mols., such as FADD, caspase-1/ICE, and caspase-3/YAMA, are present in these VSMCs, (2) that human VSMCs contain high concns. of c-FLIP (3) and that following treatment with

the protein inhibitor, CHX, cell exts. showed a decrease in c-FLIP protein that was dose- and time-dependent on the degree of apoptosis and inversely correlated with both caspase-8 and -3 activity. In contrast, there was neither a change nor an even modest upregulation of Bcl-2 family, even after 12 h of treatment with CHX. Taken together, these results may provide a novel insight into atherogenesis and suggest that c-FLIP may contribute to an apoptosis-resistant state of VSMC, and that a downregulation of c-FLIP may render VSMCs susceptible to apoptosis.

IT **169592-56-7**, Caspase-3
RL: BAC (Biological activity or effector, except adverse); BOC (Biological occurrence); BIOL (Biological study); OCCU (Occurrence)
(transition of apoptotic **resistant** vascular smooth muscle cells to troptotic sensitive state correlated with downregulation of c-FLIP)

IT **179241-78-2**, Caspase-8
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(transition of apoptotic **resistant** vascular smooth muscle cells to troptotic sensitive state correlated with downregulation of c-FLIP)

REFERENCE COUNT: 31
REFERENCE(S):
(1) Algeciras-Schimnich, A; J Immunol 1999, V162, P5205 CAPLUS
(2) Cai, W; Atherosclerosis 1997, V131, P177 CAPLUS
(4) French, L; J Cell Biol 1996, V133, P335 CAPLUS
(5) Fukuo, K; Gerontology 1997, V43, P35 CAPLUS
(6) Geng, Y; Am J Pathol 1995, V147, P251 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 3 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:878889 CAPLUS
DOCUMENT NUMBER: 134:146274
TITLE: Control of target cell survival in thyroid autoimmunity by T helper cytokines via regulation of apoptotic proteins
AUTHOR(S): Stassi, Giorgio; Di Liberto, Diana; Todaro, Matilde; Zeuner, Ann; Ricci-Vitiani, Lucia; Stoppacciaro, Antonella; Ruco, Luigi; Farina, Felicia; Zummo, Giovanni; De Maria, Ruggero
CORPORATE SOURCE: Department of Surgical, Anatomical and Oncological Sciences, Human Anatomy Section, University of Palermo, Italy
SOURCE: Nat. Immunol. (2000), 1(6), 483-488
CODEN: NIAMCZ; ISSN: 1529-2908
PUBLISHER: Nature America Inc.

DOCUMENT TYPE: Journal
LANGUAGE: English
AB After autoimmune inflammation, interactions between CD95 and its ligand (CD95L) mediate thyrocyte destruction in Hashimoto's thyroiditis (HT). Conversely, thyroid autoimmune processes that lead to Graves' disease (GD) result in autoantibody-mediated TSH receptor stimulation without thyrocyte depletion. The authors found that GD thyrocytes expressed CD95 and CD95L in a similar manner to HT thyrocytes, but did not undergo CD95-induced apoptosis either in vivo or in vitro. This pattern was due to the differential prodn. of TH1 and TH2 cytokines. Interferon .gamma. promoted caspase up-regulation and CD95-induced apoptosis in HT thyrocytes, whereas interleukin 4 and interleukin 10 protected GD thyrocytes by potent up-regulation of cFLIP and Bcl-xL, which prevented CD95-induced apoptosis in sensitized thyrocytes. Thus, modulation of apoptosis-related proteins by TH1 and TH2 cytokines controls thyrocyte survival in thyroid autoimmunity.
IT **192588-76-4**, **Proteinase** inhibitor FLIP
RL: BAC (Biological activity or effector, except adverse); BOC (Biological occurrence); BIOL (Biological study); OCCU (Occurrence)
(up-regulation in Graves' disease thyrocytes in relation to **resistance** to CD95-induced apoptosis)
REFERENCE COUNT: 37
REFERENCE(S):
(1) Abbas, A; Nature 1996, V383, P787 CAPLUS
(2) Ashkenazi, A; Science 1998, V281, P1305 CAPLUS
(3) Bretz, J; J Biol Chem 1999, V274, P25433 CAPLUS
(4) Buer, J; J Exp Med 1998, V187, P177 CAPLUS
(5) Caturegli, P; Clin Exp Immunol 1994, V98, P464 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 4 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:707290 CAPLUS
DOCUMENT NUMBER: 133:276374
TITLE: Recombinant production and biological activities of **protease-resistant** analogs of **Fas ligand** inhibitory protein
INVENTOR(S): Micanovic, Radmila; Rathnachalam, Radhakrishnan; Witcher, Derrick Ryan
PATENT ASSIGNEE(S): Eli Lilly and Co., USA
SOURCE: PCT Int. Appl., 100 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000058466	A2	20001005	WO 2000-US6418	20000320
WO 2000058466	A3	20010111		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.:

US 1999-126839	P	19990330
US 1999-140073	P	19990621
US 1999-147071	P	19990804
US 1999-160524	P	19991020
US 1999-160669	P	19991021
US 1999-172744	P	19991220
US 2000-178184	P	20000126

AB The invention relates to human **Fas ligand** inhibitory protein (FLINT) analogs that are resistant to proteolysis in vivo and in vitro at amino acid position 218 of mature FLINT, clin. and therapeutic uses thereof, and pharmaceutical formulations comprising said analogs. Analogs substituting position 218 or near this position are **resistant** to proteolysis by serine **proteinases** such as thrombin, trypsin, or chymotrypsin. Such analogs have improved half-lives and are active in inhibiting T cell activation, ischemic injury during organ transplantation, and **Fas ligand**-induced Jurkat cell apoptosis. Thus, they may have therapeutic applications for treatment of acute lung injury, acute respiratory distress syndrome, ulcerative colitis, and other disorders assocd. with abnormal apoptosis.

IT **37259-58-8**, Serine **proteinase**
RL: BAC (Biological activity or effector, except adverse); BIOL (Biological study)
(FLINT degrdn. **resistance** to; recombinant prodn. and biol. activities of **protease-resistant** analogs of **Fas ligand** inhibitory protein)

IT **210227-94-4DP**, analogs
RL: BAC (Biological activity or effector, except adverse); BPN (Biosynthetic preparation); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; recombinant prodn. and biol. activities of **protease-resistant** analogs of **Fas ligand** inhibitory protein)

L7 ANSWER 5 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:707289 CAPLUS
DOCUMENT NUMBER: 133:276373
TITLE: Recombinant production and biological activities of **Fas ligand** inhibitory factor analogs
INVENTOR(S): Becker, Gerald Wayne; Cohen, Fredric Jay; Gonzalez-Dewhitt, Patricia Ann; Hale, John Edward; Micanovic, Radmila; Newton, Christy Michelle; Noblitt, Timothy Wayne; Rathmachalam, Radhakrishnan; Tschang, Sheng-Hung Rainbow; Witcher, Derrick Ryan; Wroblewski, Victor John
PATENT ASSIGNEE(S): Eli Lilly and Co., USA
SOURCE: PCT Int. Appl., 114 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000058465	A2	20001005	WO 2000-US6417	20000320
WO 2000058465	A3	20010125		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.:
US 1999-126839 P 19990330
US 1999-140077 P 19990621
US 1999-140156 P 19990621
US 1999-160566 P 19991020
US 2000-183398 P 20000218

AB Disclosed are polypeptide analogs of human **Fas ligand** inhibitory protein (FLINT), polydeoxynucleotides encoding FLINT analogs, and methods of using FLINT analogs and polydeoxynucleotides. The FLINT analogs of the invention include polypeptides having the amino acid sequence of FLINT, modified at one or more positions with amino acid substitutions, and include fragments thereof, as well as Ig Fc region fusions comprising FLINT and FLINT analogs. Modifications in the region from about position 214 through position 222 result in **resistance** to

proteolytic cleavage by serine **pr teinases** between residues 218 and 219, resulting in analogs with longer half-lives than native FLINT. The R218Q FLINT-Fc fusion protein inhibits apoptosis. Vectors are provided for prokaryotic or mammalian cell expression and purifn. of the FLINT analogs. SUCH FLINT analogs may be useful in the treatment of disorders such as acute lung injury, acute respiratory distress syndrome, pulmonary fibrosis, chronic obstructive pulmonary disease, ulcerative colitis, Crohn's disease, and conditions assocd. with abnormal apoptosis.

IT **37259-58-8**, Serine **proteinase**
RL: BAC (Biological activity or effector, except adverse); BIOL (Biological study)
(FLINT degrdn. **resistance** to; recombinant prodn. and biol. activities of **Fas ligand** inhibitory factor analogs)

IT **210227-94-4DP**, Tumor necrosis factor receptor 6.alpha. (human precursor), analogs
RL: BAC (Biological activity or effector, except adverse); BPN (Biosynthetic preparation); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; recombinant prodn. and biol. activities of **Fas ligand** inhibitory factor analogs)

L7 ANSWER 6 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:597751 CAPLUS
DOCUMENT NUMBER: 133:265589
TITLE: Thyroid carcinoma cells are resistant to FAS-mediated apoptosis but sensitive to tumor necrosis factor-related apoptosis-inducing ligand
AUTHOR(S): Mitsiades, Nicholas; Poulaki, Vassiliki; Tseleni-Balafouta, Sophia; Koutras, Demetrios A.; Stamenkovic, Ivan
CORPORATE SOURCE: Harvard Medical School, Massachusetts General Hospital, Charlestown, MA, 02129, USA
SOURCE: Cancer Res. (2000), 60(15), 4122-4129
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Fas (APO-1/CD95) is a transmembrane protein of the tumor necrosis factor (TNF)/nerve growth factor receptor superfamily that induces apoptosis in susceptible normal and neoplastic cells upon crosslinking by its ligand (**FasL**). TNF-related apoptosis-inducing ligand (TRAIL) is a more recently identified member of the TNF superfamily that has been shown to selectively kill neoplastic cells by engaging two cell-surface receptors, DR4 and DR5. Two addnl. TRAIL receptors (DcR1 and DcR2) do not transmit

an apoptotic signal and have been proposed to confer protection from TRAIL-induced apoptosis. We addressed the expression of Fas, DR4, and DR5 in thyroid carcinoma cell lines and in 31 thyroid carcinoma specimens by Western blot anal. and immunohistochem., resp., and tested the sensitivity of thyroid carcinoma cell lines to Fas- and TRAIL-induced apoptosis. Fas was expressed in most thyroid carcinoma cell lines and tissue specimens. Although crosslinking of Fas did not induce apoptosis in thyroid carcinoma cell lines, Fas-mediated apoptosis did occur in the presence of the protein synthesis inhibitor cycloheximide, suggesting the presence of a short-lived inhibitor of the Fas pathway in these cells. Crosslinking of Fas failed to induce recruitment and activation of caspase 8, whereas transfection of a constitutively active caspase 8 construct effectively killed the SW579 papillary carcinoma cell line, arguing that the action of the putative inhibitor occurs upstream of caspase 8. By contrast, recombinant TRAIL induced apoptosis in 10 of 12 thyroid carcinoma cell lines tested, by activating caspase-10 at the receptor level and triggering a caspase-mediated apoptotic cascade. Resistance to TRAIL did not correlate with DcR1 or DcR2 protein expression and was overcome by protein synthesis inhibition in 50% of the resistant cell lines. One medullary carcinoma cell line was resistant to Fas- and TRAIL-induced apoptosis, even in the presence of cycloheximide, and to transfection of constitutively active caspase-8, suggesting a different regulation of the apoptotic pathway. These observations indicate that TRAIL effectively kills carcinomas that originate from the follicular epithelium of the thyroid gland, by inducing caspase-mediated apoptosis, and may provide a potentially potent therapeutic reagent against thyroid cancer.

IT **179241-78-2**, Caspase 8
RL: BAC (Biological activity or effector, except adverse); BIOL (Biological study)
(thyroid carcinoma cells are **resistant** to Fas-mediated apoptosis but sensitive to tumor necrosis factor-related apoptosis-inducing ligand)

REFERENCE COUNT: 57

REFERENCE(S):

(1) Arscott, P; Endocrinology 1997, V138, P5019 CAPLUS
(2) Ashkenazi, A; J Clin Investig 1999, V104, P155 CAPLUS
(3) Bellgrau, D; Nature (Lond) 1995, V377, P630 CAPLUS
(4) Beutler, B; Science (Washington DC) 1985, V229, P869 CAPLUS
(5) Blagosklonny, M; J Clin Endocrinol Metab 1998, V83, P2516 CAPLUS

ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 7 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:393482 CAPLUS
DOCUMENT NUMBER: 133:117996
TITLE: Sensitivity to Fas-mediated apoptosis is determined below receptor level in human vascular smooth muscle cells
AUTHOR(S): Chan, Shiu-Wan; Hegyi, Laszlo; Scott, Stephen; Cary, Nathaniel R. B.; Weissberg, Peter L.; Bennett, Martin R.
CORPORATE SOURCE: Unit of Cardiovascular Medicine, Addenbrooke's Centre for Clinical Investigation, Addenbrooke's Hospital, Cambridge, CB2 2QQ, UK
SOURCE: Circ. Res. (2000), 86(10), 1038-1046
CODEN: CIRUAL; ISSN: 0009-7330
PUBLISHER: Lippincott Williams & Wilkins
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Despite Fas expression, many cells resist Fas-induced apoptosis. Although differences in surface Fas expression can explain Fas resistance, multiple proteins below receptor level also inhibit Fas-induced apoptosis. To examine the mechanism of Fas resistance, we studied Fas-induced apoptosis in human medial vascular smooth muscle cells (VSMCs) from healthy coronary arteries. VSMCs showed marked heterogeneity to Fas-induced apoptosis, exhibiting both Fas-resistant (98.1 .+-. 2.3% viable, n=4, P=NS) and Fas-sensitive (31.3.+-.2.6% viable, n=3, P<0.01) cells. Fas-resistant VSMCs expressed surface Fas and could recruit RIP, indicating that functional receptor complexes were formed. However, Fas-resistant cells showed reduced expression of FADD, **Fas ligand**, and caspases 3, 7, and 8 and increased expression of FLIP and c-IAP-1. Fas-induced apoptosis was assocd. with cleavage of caspase 3 and blocked by inhibitors of caspase 3 or 8 but not caspase 1, 6, or 7. Selective inhibition of caspase 3 or 8 by antisense transfection inhibited Fas-induced apoptosis, but their reexpression could not rescue the Fas-resistant phenotype. In vivo, medial VSMCs showed marked heterogeneity of expression of caspase 3. We conclude that Fas sensitivity is detd. not only by expression of surface Fas but by differential expression of Fas-signaling proteins below receptor level. Subpopulations of cells within the same tissue have different sensitivities to apoptosis, detd. by expression of specific death-signaling proteins.

IT **169592-56-7**, Caspase 3 **179241-78-2**, Caspase 8
RL: BAC (Biological activity or effector, except adverse); BOC (Biological occurrence); BPR (Biological process); BIOL (Biological study); OCCU (Occurrence); PROC (Process)
(reduced expression of FADD, **Fas ligand**, and caspases 3, 7, and 8 and increased expression of FLIP and c-IAP-1 in Fas-**resistant** vascular smooth muscle cells)

IT **189258-14-8**, Caspase 7
RL: BOC (Biological occurrence); BPR (Biological process); BIOL (Biological study); OCCU (Occurrence); PROC (Process)
(reduced expression of FADD, **Fas ligand**, and caspases 3, 7, and 8 and increased expression of FLIP and c-IAP-1 in Fas-**resistant** vascular smooth muscle cells)

REFERENCE COUNT: 54
REFERENCE(S):
(1) Armstrong, R; J Biol Chem 1996, V271, P16850 CAPLUS
(2) Ashkenazi, A; Science 1998, V281, P1305 CAPLUS
(4) Balachandran, S; EMBO J 1998, V17, P6888 CAPLUS
(5) Bennett, M; Cardiovasc Res 1999, V41, P361 CAPLUS
(6) Bennett, M; Circ Res 1997, V81, P591 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 8 OF 21 CAPLUS COPYRIGHT 2001 ACS

ACCESSION NUMBER: 2000:384663 CAPLUS
DOCUMENT NUMBER: 133:261217
TITLE: Role of specific apoptotic pathways in the restoration of paclitaxel-induced apoptosis by valspodar in doxorubicin-resistant MCF-7 breast cancer cells
AUTHOR(S): Chadderton, Antony; Villeneuve, David J.; Gluck, Stefan; Kirwan-Rhude, Angie F.; Gannon, Brian R.; Blais, David E.; Parissenti, Amadeo M.
CORPORATE SOURCE: Department of Research, Northeastern Ontario Regional Cancer Centre, Sudbury, ON, Can.
SOURCE: Breast Cancer Res. Treat. (2000), 59(3), 231-244
CODEN: BCTRD6; ISSN: 0167-6806
PUBLISHER: Kluwer Academic Publishers
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Paclitaxel (Taxol) kills tumor cells by inducing both cellular necrosis and apoptosis. A major impediment to paclitaxel cytotoxicity is the establishment of multidrug resistance whereby exposure to one chemotherapeutic agent results in cross-resistance to a wide variety of other drugs. For example, selection of MCF-7 breast cancer cells for resistance to doxorubicin (MCF-7ADR cells) results in cross-resistance to paclitaxel. This appears to involve the overexpression of the drug transporter P-glycoprotein which can efflux both drugs from tumor cells. However, MCF-7ADR cells possess a deletion mutation in p53 and have considerably reduced levels of the Fas receptor, **Fas ligand**, caspase-2, caspase-6, and caspase-8, suggesting that paclitaxel resistance may also stem from a bona fide block in paclitaxel-induced apoptosis in

these cells. To address this issue, we examd. the ability of the P-glycoprotein inhibitor valspodar to restore paclitaxel accumulation, paclitaxel cytotoxicity, and paclitaxel-induced apoptosis. Compared to drug sensitive MCF-7 cells, MCF-7ADR cells accumulated >6-fold less paclitaxel, were approx. 100-fold more resistant to killing by the drug, and were highly resistant to paclitaxel-induced apoptosis. In contrast, MCF-7ADR cells pretreated with valspodar were indistinguishable from drug-sensitive cells in their ability to accumulate paclitaxel, in their chemosensitivity to the drug, and in their ability to undergo paclitaxel-induced apoptosis. Valspodar, by itself, did not affect these parameters. This suggests that the enhancement of paclitaxel toxicity in MCF-7ADR cells involves a restoration of apoptosis and not solely through enhanced drug-induced necrosis. Moreover, it appears that changes in the levels/activity of p53, the Fas receptor, **Fas ligand**, caspase-2, caspase-6, or caspase-8 activity have little effect on paclitaxel-induced cytotoxicity and apoptosis in human breast cancer cells.

IT **179241-78-2**, Caspase-8
RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(apoptotic pathways in the restoration of paclitaxel-induced apoptosis by valspodar in doxorubicin-**resistant** breast cancer cells)

REFERENCE COUNT: 53
REFERENCE(S):
(1) Allen, R; J Pharmacol Toxicol Meth 1997, V37, P215 CAPLUS
(3) Bachman, C; Vitro Cell Dev Biol Anim 1998, V34, P434 CAPLUS
(4) Batist, G; J Biol Chem 1986, V261, P15544 CAPLUS
(5) Beck, W; Cancer Res 1979, V39, P2070 CAPLUS
(7) Cai, Z; Int J Cancer 1996, V68, P535 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 9 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:285406 CAPLUS
DOCUMENT NUMBER: 133:72160
TITLE: Resistance to CD95/Fas-induced and ceramide-mediated apoptosis of human melanoma cells is caused by a defective mitochondrial cytochrome c release
AUTHOR(S): Raisova, M.; Bektas, M.; Wieder, T.; Daniel, P.; Eberle, J.; Orfanos, C. E.; Geilen, C. C.
CORPORATE SOURCE: University Medical Center Benjamin Franklin, Department of Dermatology, The Free University of Berlin, Berlin, Germany
SOURCE: FEBS Lett. (2000), 473(1), 27-32

CODEN: FEBLAL; ISSN: 0014-5793
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Intracellular CD95/Fas-signaling pathways have not been investigated in melanoma yet. Two different CD95 receptor-induced apoptotic pathways are presently known in other cell types: (i) direct activation of caspase-8 and (ii) induction of ceramide-mediated mitochondrial activation, both leading to subsequent caspase-3 activation. In the present study, five of 11 melanoma cell populations were shown to release cytochrome c from mitochondria, which activates caspase-3 and finally results in DNA fragmentation upon treatment with the agonistic monoclonal antibody CH-11. In contrast, this apoptotic pathway was not activated in the remaining six melanoma cell populations. Interestingly, the susceptibility of melanoma cells to CD95L/**FasL**-triggered cell death was clearly correlated with N-acetylsphingosine-mediated apoptosis. Our results are in line with a defect upstream of mitochondrial cytochrome c release in resistant cells.

IT **169592-56-7**, Caspase 3
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(activation in apoptotic pathway of human melanoma cells in relation to **resistance** to CD95/Fas-induced and ceramide-mediated apoptosis caused by defective mitochondrial cytochrome c release)

REFERENCE COUNT: 41
REFERENCE(S):
(1) Adams, J; Science 1998, V281, P1322 CAPLUS
(2) Anjum, R; FEBS Lett 1998, V439, P81 CAPLUS
(3) Ashkenazi, A; Science 1998, V281, P1305 CAPLUS
(6) Bargou, R; J Clin Invest 1996, V97, P2651 CAPLUS
(8) Boldin, M; Cell 1996, V85, P803 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 10 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:213161 CAPLUS
DOCUMENT NUMBER: 132:342934
TITLE: HMBA induces activation of a caspase-independent cell death pathway to overcome P-glycoprotein-mediated multidrug resistance
AUTHOR(S): Ruefli, Astrid A.; Smyth, Mark J.; Johnstone, Ricky W.
CORPORATE SOURCE: Austin Research Institute, Austin Hospital, Heidelberg, 3084, Australia
SOURCE: Blood (2000), 95(7), 2378-2385
CODEN: BLOOAW; ISSN: 0006-4971

PUBLISHER: American Society of Hematology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Multidrug resistance (MDR) is often characterized by the expression of P-glycoprotein (P-gp), a 170 kDa ATP-dependent drug efflux protein. As well as effluxing xenotoxins, functional P-gp can confer resistance to caspase-dependent apoptosis induced by a range of different stimuli, including **Fas ligand**, tumor necrosis factor, UV irradn., and serum starvation. However, P-gp-pos. cells remain sensitive to caspase-independent death induced by cytotoxic T-cell granule proteins, perforin, and granzyme B. It is, therefore, possible that agents that induce cell death in a caspase-independent manner might circumvent P-gp-mediated MDR. The authors demonstrated here that hexamethylene bisacetamide (HMBA) induced equiv. caspase-independent cell death in both P-gp-pos. and -neg. cell lines at concns. of 10 mmol/L and above. The HMBA-induced death pathway was marked by release of cytochrome c from the mitochondria and redn. of Bcl-2 protein levels. In addn., the authors show that functional P-gp specifically inhibits the activation of particular caspases, such as caspases-8 and -3, whereas others, such as caspase-9, remain unaffected. These studies greatly enhance the authors understanding of the mol. cell death events that can be regulated by functional P-gp and highlight the potential clin. use of drugs that function via a caspase-independent pathway for the treatment of MDR tumors.
IT **169592-56-7**, Caspase-3 **179241-78-2**, Caspase-8
RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(HMBA induces activation of a caspase-independent cell death pathway in tumor cells to overcome P-glycoprotein-mediated multidrug **resistance** in relation to mechanism)
REFERENCE COUNT: 47
REFERENCE(S):
(2) Boldin, M; Cell 1996, V85, P803 CAPLUS
(4) Bradford, M; Anal Biochem 1976, V72, P248 CAPLUS
(5) Broggini, M; Biochem Pharmacol 1988, V37, P4423 CAPLUS
(6) Byrd, J; Blood 1998, V92, P3804 CAPLUS
(7) de Maria, R; Science 1997, V277, P1652 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 11 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:736804 CAPLUS
DOCUMENT NUMBER: 132:48979
TITLE: Dendritic cells are resistant to apoptosis through the fas (CD95/APO-1) pathway
AUTHOR(S): Ashany, Dalit; Savir, Asaf; Bhardwaj, Nina;

Elkon, Keith B.
CORPORATE SOURCE: Hospital for Special Surgery, Cornell University Medical Center, New York, NY, 10021, USA
SOURCE: J. Immunol. (1999), 163(10), 5303-5311
CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Immunoregulation of lymphocytes and macrophages in the peripheral immune system is achieved in part by activation-induced cell death. Members of the TNF receptor family including Fas (CD95) are involved in the regulation of activation-induced cell death. To det. whether activation-induced cell death plays a role in regulation of dendritic cells (DCs), we examd. interactions between Ag-presenting murine DCs and Ag-specific Th1 CD4+ T cells. Whereas mature bone marrow- or spleen-derived DCs expressed high levels of Fas, these DCs were relatively insensitive to Fas-mediated killing by the agonist mAb, Jo-2, as well as authentic **Fas ligand** expressed on the CD4+ T cell line, A.E7. The insensitivity to Fas-mediated apoptosis was not affected by priming with IFN-.gamma. and/or TNF-.alpha. or by blocking the DC survival signals TNF-related activation-induced cytokine and CD40L. However, apoptosis could be induced with C2-ceramide, suggesting that signals proximal to the generation of ceramide might mediate resistance to Fas. Anal. of protein expression of several anti-apoptotic mediators revealed that expression of the intracellular inhibitor of apoptosis Fas-assocd. death domain-like IL-1-converting enzyme-inhibitory protein was significantly higher in Fas-resistant DCs than in Fas-sensitive macrophages, suggesting a possible role for Fas-assocd. death domain-like IL-1-converting enzyme-inhibitory protein in DC resistance to Fas-mediated apoptosis. Our results demonstrate that murine DCs differ significantly from other APC populations in susceptibility to Fas-mediated apoptosis during cognate presentation of Ag. Because DCs are most notable for initiation of an immune response, resistance to apoptosis may contribute to this function.
IT **179241-78-2**
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(dendritic cells are **resistant** to apoptosis through the Fas (CD95/APO-1) pathway and)
REFERENCE COUNT: 55
REFERENCE(S):
(2) Albert, M; Nature 1998, V392, P86 CAPLUS
(3) Anderson, D; Nature 1997, V390, P175 CAPLUS
(4) Ashany, D; Proc Natl Acad Sci USA 1995, V92, P11225 CAPLUS
(5) Banchereau, J; Nature 1998, V392, P245 CAPLUS

(7) Bjorck, P; Int Immunol 1997, V9, P365 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 12 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:319630 CAPLUS
DOCUMENT NUMBER: 131:143472
TITLE: Blockade of the Fas-triggered intracellular signaling pathway in human melanomas is circumvented by cytotoxic lymphocytes
AUTHOR(S): Ferrarini, Marina; Imro, Maria Adele; Sciorati, Clara; Heltai, Silvia; Protti, Maria Pia; Pellicciari, Carlo; Rovere, Patrizia; Manfredi, Angelo A.; Rugarli, Claudio
CORPORATE SOURCE: Laboratorio di Immunologia dei Tumori, Divisione di Medicina II, H San Raffaele Scientific Institute, Milan, Italy
SOURCE: Int. J. Cancer (1999), 81(4), 573-579
CODEN: IJCNAW; ISSN: 0020-7136
PUBLISHER: Wiley-Liss, Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Fas and **Fas ligand** (**FasL**) have been found both in lymphoid and in non-lymphoid malignancies, and are thought to play a role in the interplay between tumors and the immune system. Here the authors investigated Fas/**FasL** expression, function, and intracellular signaling pathways in human melanomas. Of 5 melanoma cell lines, 3 expressed Fas at their surface, and all of them expressed **FasL**. **FasL** was functional, since it triggered Fas-induced apoptosis of human T cell clones. Conversely, crosslinking of Fas mol. with a specific monoclonal antibody failed to induce apoptosis in any of the melanomas tested, or ceramide intracellular accumulation or caspase-3 activation, pointing to an early alteration in the Fas-triggered signaling cascade. All melanomas retained the ability to undergo apoptosis induced by cytotoxic lymphocytes, which was mediated by the granule exocytosis mechanism. This suggests that melanoma cells evade immune-mediated Fas-triggered apoptosis via a selective blockade of the Fas apoptotic pathway. Cytotoxic lymphocytes, however, may circumvent tumor resistance to Fas-induced death via granzyme-mediated apoptosis, further supporting the development of immunotherapeutic strategies in the treatment of cancer.

IT **143180-74-9**, Granzyme B
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(Fas/**FasL**-mediated apoptosis **resistance** in human melanomas is circumvented by interleukin-2-activated cytotoxic lymphocytes via granule exocytosis)

REFERENCE COUNT: 21
REFERENCE(S):
(1) Alderson, M; J exp Med 1995, V181, P71 CAPLUS
(2) Andrade, F; Immunity 1998, V8, P451 CAPLUS
(3) Atkinson, E; Crit Rev Immunol 1995, V15, P359 CAPLUS
(4) Buechner, S; J clin Invest 1997, V100, P2691 CAPLUS
(6) Enari, M; Nature (Lond) 1996, V380, P723 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 13 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:290905 CAPLUS
DOCUMENT NUMBER: 131:139046
TITLE: Selective inhibition of apoptosis by TPA-induced differentiation of U937 leukemic cells
AUTHOR(S): Sordet, Olivier; Bettaieb, Ali; Bruey, Jean-Marie; Eymin, Beatrice; Droin, Nathalie; Ivarsson, Michael; Garrido, Carmen; Solary, Eric
CORPORATE SOURCE: Department of Biology and Therapy of Cancer, INSERM U517, JE515, Faculty of Medicine and Pharmacy, Dijon, 21033, Fr.
SOURCE: Cell Death Differ. (1999), 6(4), 351-361
CODEN: CDDIEK; ISSN: 1350-9047
PUBLISHER: Stockton Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB U937 leukemic cells treated for 24 h with 16 nM 12-O-tetradecanoylphorbol 13-acetate (TPA), that induces their macrophagic terminal differentiation, become resistant to etoposide-induced apoptosis. Exposure of undifferentiated U937 cells to 50 .mu.M etoposide for 6 h, that triggers apoptosis in 80% cells, activates procaspase-2L, -3 and -8, induces the mitochondrial release of cytochrome c and decreases Mcl-1 expression without modifying Bcl-2, Bcl-xL and Bax protein levels. All these events are inhibited in TPA-differentiated U937 cells that are also resistant to vinblastine-induced and Fas-mediated cell death. Interestingly, these cells are not inherently resistant to apoptosis induction. Exposure of TPA-differentiated U937 cells to 0.8 .mu.g/mL cycloheximide for 24 h, that triggers apoptosis in 50% cells, activates procaspase-2L, -3 and -8, induces the mitochondrial release of cytochrome c and decreases Bcl-xL expression without modifying Bcl-2, Mcl-1 and Bax protein levels. All these events are not obsd. in undifferentiated cells treated in similar conditions. These results indicate that the apoptotic pathway that involves the release of cytochrome c from mitochondria and the cleavage of procaspases remains functional in TPA-differentiated cells.

IT **169592-56-7**, Caspase-3
RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(TPA-induced differentiation of leukemic cells and **resistance** to etoposide-induced apoptosis)

REFERENCE COUNT: 62
REFERENCE(S):
(1) Aihara, H; Proc Natl Acad Sci USA 1991, V88, P11062 CAPLUS
(2) Bertrand, R; Drug Develop Res 1995, V34, P138 CAPLUS
(3) Boudreau, N; Science 1995, V267, P891 CAPLUS
(4) Cardone, M; Cell 1997, V90, P315 CAPLUS
(5) Chatterjee, D; Cell Growth Differ 1997, V8, P1083 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 14 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:270721 CAPLUS
DOCUMENT NUMBER: 131:42586
TITLE: Mitochondrial regulation of cell death: mitochondria are essential for procaspase 3-p21 complex formation to resist Fas-mediated cell death
AUTHOR(S): Suzuki, Atsushi; Tsutomi, Yumi; Yamamoto, Naoe; Shibutani, Tomoko; Akahane, Kouichi
CORPORATE SOURCE: Drug Safety Research Laboratory, Project for the Cell Death Research, Daiichi Pharmaceutical Co., Ltd., Tokyo, 134-8630, Japan
SOURCE: Mol. Cell. Biol. (1999), 19(5), 3842-3847
CODEN: MCEBD4; ISSN: 0270-7306
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Death receptor Fas transduces cell death signaling upon stimulation by **Fas ligand**, and this death signaling is mediated by caspase. Recently, we reported that the cell cycle regulator p21 interacts with procaspase 3 to resist Fas-mediated cell death. In the present study, the mol. characterization and functional region of the procaspase 3-p21 complex was further investigated. We obsd. the p21 expression in the mitochondrial fraction of HepG2 cells and detected Fas-mediated cell death only in the presence of actinomycin D. However, mitochondrial-DNA-lacking HepG2 (MDLH) cells showed this effect even in the absence of actinomycin D. Both p21 and procaspase 3 were expressed in MDLH cells, but the procaspase 3-p21 complex formation was not obsd. Interestingly, the resistance to Fas-mediated cell death in the MDLH cells without actinomycin D was recovered after microinjection of HepG2-derived mitochondria into the MDLH cells. We conclude that

mitochondria are necessary for procaspase 3-p21 complex formation and propose that the mitochondrial role during cell death is not only death induction but also death suppression.

IT **169592-56-7**, Caspase 3
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(mitochondrial regulation of cell death: mitochondria are essential for procaspase 3-p21 complex formation to **resist** Fas-mediated cell death)

REFERENCE COUNT: 42
REFERENCE(S):
(1) Alnemri, E; Cell 1996, V87, P171 CAPLUS
(2) Boldin, M; Cell 1996, V85, P803 CAPLUS
(3) Chen, J; Nature 1995, V374, P386 CAPLUS
(4) Darmon, A; Nature 1995, V377, P446 CAPLUS
(5) Desjardins, P; Mol Cell Biol 1985, V5, P1163 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 15 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:220052 CAPLUS
DOCUMENT NUMBER: 130:266383
TITLE: Novel human **Fas ligand** (**FasL**) derivatives with improved **proteinase-resistance** for use as apoptosis modulators
INVENTOR(S): Nagata, Shigekazu; Tanaka, Masato
PATENT ASSIGNEE(S): Mochida Pharmaceutical Co., Ltd., Japan; Osaka Bioscience Institute
SOURCE: PCT Int. Appl., 60 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9914325	A1	19990325	WO 1998-JP4187	19980917
W: CA, JP, US				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
EP 1016721	A1	20000705	EP 1998-943025	19980917
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				

PRIORITY APPLN. INFO.: JP 1997-252541 A 19970917
WO 1998-JP4187 W 19980917

AB A novel sol., **proteinase-resistant FasL** functioning as a Fas antagonist or apoptosis modulator is

derivatized from human **FasL**;. The deriv. is particularly resistant to metalloproteinase. The **FasL** deriv., exhibiting an excellent apoptosis-inducing activity, is prepd. by deleting/substituting the natural human **FasL** in the regions of 1.apprx.129/130, 111.apprx.128, 131.apprx.133, and optionally, by deleting the region of 8.apprx.69. The.

IT **9001-92-7, Proteinase**
RL: ADV (Adverse effect, including toxicity); BIOL (Biological study)
(novel human **Fas ligand** (**FasL**) derivs. with improved **proteinase-resistance** for use as apoptosis modulators)

REFERENCE COUNT: 4
REFERENCE(S):
(1) Sumitomo Electric Industries Ltd; JP 09124510 A 1997 CAPLUS
(2) Takahashi, T; International Immunology 1994, V6(10), P1567 CAPLUS
(3) Tanaka, M; Nature Medicine 1996, V2(3), P317 CAPLUS
(4) Tanaka, M; Nature Medicine 1998, V4(5), P31

L7 ANSWER 16 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:194259 CAPLUS
DOCUMENT NUMBER: 130:233258
TITLE: Viral vector system capable of expressing an apoptosis-associated gene
INVENTOR(S): Hamada, Hirofumi
PATENT ASSIGNEE(S): RPR Gencell Asia/Pacific Inc., Japan
SOURCE: PCT Int. Appl., 51 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9913073	A2	19990318	WO 1998-JP4010	19980907
WO 9913073	A3	19990610		
W: AU, CA, KR, NZ, US				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
JP 11075859	A2	19990323	JP 1997-259235	19970908
AU 9889991	A1	19990329	AU 1998-89991	19980907

PRIORITY APPLN. INFO.: JP 1997-259235 19970908
WO 1998-JP4010 19980907

AB An apoptosis-resistant virus-sensitive cell line based upon cell line 293 is disclosed. To such cells, apoptosis resistance genes

such as crmA, bcl-2, bcl-x1, FLIP, survivin, IAP, or ILP have been introduced. The generation of adenovirus vectors capable of expressing apoptosis-assocd. genes such as FAS, FLICE, bcl-xs, and Bax is achieved using said cell line. The recombinant viruses of the invention may be useful for gene therapy for cancer, autoimmune diseases, graft rejection, and inflammatory diseases.

L7 ANSWER 17 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:190562 CAPLUS
DOCUMENT NUMBER: 130:351011
TITLE: Cytokine-based tumor cell vaccine is equally effective against parental and isogenic multidrug-resistant myeloma cells: the role of cytotoxic T lymphocytes
AUTHOR(S): Shtil, Alexander A.; Turner, Joel G.; Durfee, John; Dalton, William S.; Yu, Hua
CORPORATE SOURCE: Clinical Investigations Program and Immunology Program, H. Lee Moffitt Cancer Center and Research Institute, Tampa, FL, 33612-9497, USA
SOURCE: Blood (1999), 93(6), 1831-1837
CODEN: BLOOAW; ISSN: 0006-4971
PUBLISHER: W. B. Saunders Co.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Tumor cells that survive initial courses of chemotherapy may do so by acquiring a multidrug-resistant phenotype. This particular mechanism of drug resistance may also confer resistance to physiol. effectors of apoptosis that could potentially reduce the efficacy of immune therapies that use these pathways of cell death. We have previously demonstrated high efficacy for a cytokine-based tumor cell vaccine in a murine MPC11 myeloma model. In the present study, the effects of this vaccination were compared in MPC11 cells and their isogenic sublines selected for mdr1/P-glycoprotein (Pgp)-mediated multidrug resistance (MDR). Immunization with MPC11 cells expressing granulocyte-macrophage colony-stimulating factor (GM-CSF) and interleukin-12 (IL-12) led to long-lasting protection of mice against s.c. challenge with both parental cells or their MDR variants. Similarly, immunization with GM-CSF/IL-12-transfected MDR sublines caused rejection of transplantation of both parental cells and the MDR sublines. Whereas MPC11 cells and their MDR variants were resistant to APO-1/CD95/**Fas ligand**, the immunization generated potent granzyme B/perforin-secreting cytotoxic T lymphocytes (CTLs) that were similarly effective against both parental and isogenic MDR cells. We conclude that MDR mediated by mdr1/Pgp did not interfere with lysis by pore-forming CTLs. Immunotherapy based on pore-forming CTLs may be an attractive approach to the treatment of drug-resistant myeloma.

IT **143180-74-9**, Granzyme B

RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(cytokine-based tumor cell vaccine is equally effective against parental and isogenic multidrug-**resistant** myeloma cells and is mediated by granzyme B/perforin-secreting cytotoxic T-cells)

REFERENCE COUNT: 40

REFERENCE(S):
(1) Atkinson, E; J Biol Chem 1998, V273, P21261 CAPLUS
(2) Berke, G; Annu Rev Immunol 1994, V12, P735 CAPLUS
(3) Berke, G; Cell 1995, V81, P9 CAPLUS
(4) Dalton, W; Blood 1989, V73, P747 CAPLUS
(5) Dranoff, G; Proc Natl Acad Sci USA 1993, V90, P3539 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 18 OF 21 CAPLUS COPYRIGHT 2001 ACS

ACCESSION NUMBER: 1999:7108 CAPLUS

DOCUMENT NUMBER: 130:221254

TITLE: Anti-Fas antibody differentially regulates apoptosis in **Fas ligand** **resistant** carcinoma lines via the caspase 3 family of cell death **proteases** but independently of bcl2 expression

AUTHOR(S): Crowe, David L.; Boardman, Mitzi L.; Fong, Kimberlee S.

CORPORATE SOURCE: Center for Craniofacial Molecular Biology, University of Southern California, Los Angeles, CA, 90033, USA

SOURCE: Anticancer Res. (1998), 18(5A), 3163-3170
CODEN: ANTRD4; ISSN: 0250-7005

PUBLISHER: Anticancer Research

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Deregulation of cell death pathways is an important feature of tumorigenesis. Fas, a member of the tumor necrosis factor receptor superfamily, is a transmembrane protein that can transduce cell death signals via a proteolytic cascade upon crosslinking or ligand binding. Fas has been implicated in the cell turnover of normal stratified squamous epithelia. To det. if altered Fas mediated cell death pathways participate in epithelial tumorigenesis, the authors examd. squamous cell carcinoma (SCC) lines for sensitivity to **Fas ligand** (**FasL**) or an agonistic anti-Fas antibody. All cell lines examd. were resistant to **FasL** mediated cell death. The carcinoma cell line SCC71 was also highly resistant to anti-Fas antibody. Another line, SCC9, underwent rapid cell death with characteristic features of apoptosis

after exposure to anti-Fas antibody. However, binding of both **FasL** and anti-Fas antibody recruited downstream effector mols. to the Fas cytoplasmic domain in both SCC9 and SCC71 cells. Inhibition of the caspase 3 but not the ICE family of cell death proteases blocked apoptosis in SCC9 cells independently of expression of the anti-apoptotic protein bcl2. Thus, Fas differentially mediates apoptosis in SCC lines by activation of caspase 3 family members but independent of bcl2 expression.

REFERENCE COUNT: 38

REFERENCE(S):
(1) Alnemri, E; Cell 1996, V87, P171 CAPLUS
(2) Atkinson, E; J Biol Chem 1996, V271, P5968 CAPLUS
(3) Bellgrau, D; Nature 1995, V377, P630 CAPLUS
(4) Boldin, M; Cell 1996, V85, P803 CAPLUS
(5) Brinkmann, U; Proc Natl Acad Sci USA 1995, V92, P10427 CAPLUS

ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 19 OF 21 CAPLUS COPYRIGHT 2001 ACS

ACCESSION NUMBER: 1998:419703 CAPLUS
DOCUMENT NUMBER: 129:197640

TITLE: Deficient activation of the CD95 (APO-1/Fas) system in drug-resistant cells

AUTHOR(S): Friesen, C.; Fulda, S.; Debatin, K. -M.
CORPORATE SOURCE: German Cancer Research Center, Hematology/Oncology, University Children's Hospital, Ulm, D-89075, Germany

SOURCE: Leukemia (1997), 11(11), 1833-1841
CODEN: LEUKED; ISSN: 0887-6924
PUBLISHER: Stockton Press
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The mol. mechanisms for sensitivity and resistance of tumor cells towards chemotherapy are only partially understood. In chemosensitive leukemias and solid tumors, anticancer drugs have been shown to induce apoptosis. The authors previously identified activation of the CD95 (APO-1/Fas) receptor/CD95 ligand (CD95/CD95-L) system as a key mechanism for drug-induced apoptosis. Here, the authors show that therapeutic concns. of doxorubicin, methotrexate and cytarabine also induce apoptosis via activation of the CD95 system in primary leukemia cells in vivo. CD95-resistant and doxorubicin-resistant leukemia and neuroblastoma cells display cross-resistance for induction of cell death. Down-regulation of CD95 expression was found in drug-resistant and CD95-resistant cell lines. Furthermore, up-regulation of CD95-L, previously shown to mediate drug-induced apoptosis in a variety of tumor cells, was completely blocked in doxorubicin-resistant cells. The prototype caspase (ICE/Ced-3 **protease**) substrate,

poly(ADP-ribose)polymerase (PARP), was cleaved in sensitive, but not in **resistant** tumor cells following CD95 triggering or drug treatment. Since failure to activate CD95-L was not due to decreased drug uptake or increased drug efflux, non-multi-drug resistance (non-MDR) mechanisms are involved in this type of resistance. These findings suggested that an intact CD95 system plays a key role in detg. sensitivity or resistance towards anticancer therapy.

L7 ANSWER 20 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:95628 CAPLUS
DOCUMENT NUMBER: 128:178792
TITLE: Resistance to apoptosis correlates with a highly proliferative phenotype and loss of Fas and CPP32 (caspase-3) expression in human leukemia cells
AUTHOR(S): Martinez-Lorenzo, Maria J.; Gamen, Susana; Etxeberria, Jaime; Lasierra, Pilar; Larrad, Luis; Pineiro, Andres; Anel, Alberto; Naval, Javier; Alava, Maria A.
CORPORATE SOURCE: Departamento de Bioqu(mica y Biologia Molecular y Celular, Facultad de Ciencias, Universidad de Zaragoza, Zaragoza, 50009, Spain
SOURCE: Int. J. Cancer (1998), 75(3), 473-481
CODEN: IJCNAW; ISSN: 0020-7136
PUBLISHER: Wiley-Liss, Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Apoptosis induced by effector cells of the immune system or by cytotoxic drugs is a main mechanism mediating the prevention or elimination of tumoral cells. For instance, the human T-cell leukemia Jurkat is sensitive to Fas-induced apoptosis and to activation-induced cell death (AICD), and the promonocytic leukemia U937 is sensitive to Fas- and TNF-induced apoptosis. In this work, the authors have analyzed the mechanisms of resistance to physiol. or pharmacol. apoptosis in human leukemia by generating highly proliferative (hp) sub-lines derived from Jurkat and U937 cells. These hp sub-lines were resistant to Fas- and TNF-induced apoptosis, as well as to AICD. This was due to the complete loss of Fas and TNFR surface expression and, in the case of Jurkat-derived sub-lines, also of CD3, CD2 and CD59 mols. The sub-lines also completely lacked the expression of the apoptotic protease CPP32, present in parental cells. Moreover, these sub-lines were no longer sensitive to doxorubicin-induced apoptosis, which was efficiently blocked by the general caspase inhibitor Z-VAD-fmk in the parental cell lines. These data suggest a mol. mechanism for the development of resistance of leukemic cells to physiol. and pharmacol. apoptosis inducers, giving rise to highly proliferative tumoral phenotypes.

These results also indicate that Fas and CPP32 could be useful prognostic markers for the progression and/or therapy outcome of human leukemias.

IT **169592-56-7**, Caspase-3
RL: BOC (Biological occurrence); BPR (Biological process); BIOL (Biological study); OCCU (Occurrence); PROC (Process)
(**resistance** to apoptosis correlates with a highly proliferative phenotype and loss of Fas and CPP32 (caspase-3) expression in human leukemia cells)

L7 ANSWER 21 OF 21 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:729416 CAPLUS
DOCUMENT NUMBER: 128:43516
TITLE: Betulinic acid triggers CD95 (APO-1/Fas)- and p53-independent apoptosis via activation of caspases in neuroectodermal tumors
AUTHOR(S): Fulda, Simone; Friesen, Claudia; Los, Marek; Scaffidi, Carsten; Mier, Walter; Benedict, Mary; Nunez, Gabriel; Krammer, Peter H.; Peter, Marcus E.; Debatin, Klaus-Michael
CORPORATE SOURCE: Division of Hematology/Oncology, German Cancer Research Center, University Children's Hospital and Division of Molecular Oncology, Heidelberg, D-69120, Germany
SOURCE: Cancer Res. (1997), 57(21), 4956-4964
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Betulinic acid (BA), a melanoma-specific cytotoxic agent, induced apoptosis in neuroectodermal tumors, such as neuroblastoma, medulloblastoma, and Ewing's sarcoma, representing the most common solid tumors of childhood. BA triggered an apoptosis pathway different from the one previously identified for std. chemotherapeutic drugs. BA-induced apoptosis was independent of CD95-ligand/receptor interaction and accumulation of wild-type p53 protein, but it critically depended on activation of caspases (interleukin 1.beta.-converting enzyme/Ced-3-like proteases). FLICE/MACH (caspase-8), considered to be an upstream protease in the caspase cascade, and the downstream caspase CPP32/YAMA/Apopain (caspase-3) were activated, resulting in cleavage of the prototype substrate of caspases PARP. The broad-spectrum peptide inhibitor benzyloxycarbonyl-Val-Ala-Asp-fluoromethylketone, which blocked cleavage of FLICE and PARP, also completely abrogated BA-triggered apoptosis. Cleavage of caspases was preceded by disturbance of mitochondrial membrane potential and by generation of reactive oxygen species. Overexpression of Bcl-2 and Bcl-xL conferred **resistance** to BA at the level of mitochondrial dysfunction,

protease activation, and nuclear fragmentation. This suggested that mitochondrial alterations were involved in BA-induced activation of caspases. Furthermore, Bax and Bcl-xs, two death-promoting proteins of the Bcl-2 family, were up-regulated following BA treatment. Most importantly, neuroblastoma cells resistant to CD95- and doxorubicin-mediated apoptosis were sensitive to treatment with BA, suggesting that BA may bypass some forms of drug resistance. Because BA exhibited significant antitumor activity on patients' derived neuroblastoma cells ex vivo, BA may be a promising new agent for the treatment of neuroectodermal tumors in vivo.

IT **169592-56-7**, Caspase 3 **179241-78-2**, Caspase 8
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(betulinic acid triggers CD95 APO-1/Fas- and p53-independent apoptosis via activation of caspases in neuroectodermal tumors in relation to Bcl-2 family proteins expression and mitochondrial dysfunction and **resistance**)

(FILE 'MEDLINE, BIOSIS, EMBASE, WPIDS, CONFSCI, SCISEARCH, JICST-EPLUS, JAPIO' ENTERED AT 10:37:49 ON 26 APR 2001)

L8 69 S L7
L9 31 DUP REM L8 (38 DUPLICATES REMOVED)

L9 ANSWER 1 OF 31 MEDLINE DUPLICATE 1

ACCESSION NUMBER:	2001151187 MEDLINE
DOCUMENT NUMBER:	21115851 PubMed ID: 11221844
TITLE:	Signaling events triggered by tumor necrosis factor-related apoptosis-inducing ligand (TRAIL): caspase-8 is required for TRAIL-induced apoptosis.
AUTHOR:	Seol D W; Li J; Seol M H; Park S Y; Talanian R V; Billiar T R
CORPORATE SOURCE:	Department of Surgery, University of Pittsburgh School of Medicine, Pennsylvania 15261 USA.. seold+@pitt.edu
CONTRACT NUMBER:	GM44100 (NIGMS) GM53789 (NIGMS)
SOURCE:	CANCER RESEARCH, (2001 Feb 1) 61 (3) 1138-43. Journal code: CNF. ISSN: 0008-5472.
PUB. COUNTRY:	United States Journal; Article; (JOURNAL ARTICLE)
LANGUAGE:	English
FILE SEGMENT:	Priority Journals
ENTRY MONTH:	200103
ED	Entered STN: 20010404 Last Updated on STN: 20010404 Entered PubMed: 20010226 Entered Medline: 20010315

AB Tumor necrosis factor (TNF)-related apoptosis-inducing ligand (TRAIL) is a TNF family member and potent apoptosis inducer. In contrast to TNF-alpha or **Fas ligand**, relatively little is known about the signaling events activated by TRAIL. In particular, the initial caspase(s) required for TRAIL-induced apoptosis remains to be determined Caspase-3-like **protease** but not caspase-1-like **protease** (YVADase) activity rapidly increased in HeLa cells in response to TRAIL treatment. The increase in **protease** activity correlated with the profile of apoptotic cell death that was inhibited by the pan-caspase inhibitor Z-VAD-fmk. In response to TRAIL, caspase-8, an initiator caspase in death receptor-mediated apoptosis, was activated within 1 h in association with Bid cleavage, cytochrome c release, caspase-3 activation, and DNA fragmentation factor 45 cleavage. Z-IETD-fmk, a caspase-8 inhibitor, completely blocked caspase-8 activation and resulted in inhibition of caspase-3 (a caspase-3-like **protease**) activation and apoptotic cell death. Overexpression of a caspase-8 dominant negative mutant inhibited apoptosis induced by TRAIL. Caspase-8-deficient Jurkat cells were **resistant** to both TRAIL and Fas-induced apoptosis, whereas wild-type Jurkat cells were susceptible to both TRAIL- and Fas-induced apoptosis. The caspase-8-reintro duced caspase-8-deficient Jurkat cells acquired normal susceptibility to both TRAIL and agonistic Fas antibody. Reverse transcription-PCR and sequence analyses have revealed that these caspase-8-deficient Jurkat cell express wild-type caspase-10. Therefore, our data indicate that caspase-8 is required for TRAIL-induced apoptosis and suggest that caspase-10 may play a minor role, if any, in TRAIL-induced apoptosis.

L9 ANSWER 2 OF 31 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.

ACCESSION NUMBER: 2001047083 EMBASE

TITLE: Mutation analysis of the FAS and TNFR apoptotic cascade genes in hematological malignancies.

AUTHOR: Rozenfeld-Granot G.; Toren A.; Amariglio N.; Brok-Simoni F.; Rechavi G.

CORPORATE SOURCE: G. Rozenfeld-Granot, Institute of Hematology, Chaim Sheba Medical Center, Tel-Hashomer 52621, Israel. galitg@post.tau.ac.il

SOURCE: Experimental Hematology, (2001) 29/2 (228-233). Refs: 38 ISSN: 0301-472X CODEN: EXHEBH

PUBLISHER IDENT.: S 0301-472X(00)00623-8

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 022 Human Genetics
025 Hematology

LANGUAGE: English

SUMMARY LANGUAGE: English

AB Objective - The existence of properly functioning apoptotic pathways is of utmost importance in the maintenance of a normal cell count. Several groups have searched for mutations in the FAS receptor, a well-characterized apoptotic protein carrying a death domain, and reported the existence of rare mutations in multiple myeloma, T-acute lymphoblastic leukemia (T-ALL), and adult T-cell leukemia. Our aim was to expand these searches by looking for mutations in the death domains of FAS, FADD, TNFR, TRADD, and RIP, in the promoter region of FAS, and in the **protease** domain of caspase 10, in a larger variety of hematological malignancies, some of which express an apoptosis-**resistant** phenotype. Methods - We extracted RNA and DNA samples from 92 hematological malignancies: chronic lymphocytic leukemia (CLL; 31 cases), chronic myelogenous leukemia (CML; 28 cases), essential thrombocythemia (ET; 8 cases), acute lymphocytic leukemia (ALL; 6 cases), acute myeloblastic leukemia (AML; 6 cases), hairy-cell leukemia (HCL; 3 cases), Burkitt's lymphoma (3 cases), polycythemia vera (PV; 3 cases), myelofibrosis (2 cases), and chronic myelomonocytic leukemia (CMML; 2 cases) and performed PCR-SSCP and sequence analysis on these samples. Results - Five polymorphic patterns were found: three in the death domain of the FAS gene in CML patients, one in the promoter of this gene in a CLL patient, and the fifth in the death domain of the TRADD gene in a CML patient. No mutations, altering amino acids, were found in these genes in any of the aforementioned malignancies. Conclusions - These observations imply that mutations in the death domains of FAS, FADD, TNFR, TRADD, and RIP and in the **protease** domain of caspase 10 are not a major cause for failure of apoptosis in hematological malignancies, mainly CML and CLL. Regulatory and epigenetic abnormalities in these apoptotic cascade members and aberrations in other components of all death machinery should be looked for. Copyright .COPYRGT. 2001 International Society for Experimental Hematology.

L9 ANSWER 3 OF 31 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD

ACCESSION NUMBER: 2000-664925 [64] WPIDS
CROSS REFERENCE: 2000-656167 [60]
DOC. NO. CPI: C2000-201388
TITLE: Novel **protease resistant** **FAS ligand** inhibitory protein analogs **resistant** to in vivo or in vitro proteolysis at amino acid position 218 of the mature protein, useful for treating autoimmune diseases.
DERWENT CLASS: B04 D16
INVENTOR(S): MICANOVIC, R; RATHNACHALAM, R; WITCHER, D R
PATENT ASSIGNEE(S): (ELIL) LILLY & CO ELI
COUNTRY COUNT: 91

PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000058466	A2	20001005	(200064)*	EN	100

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SL SZ TZ UG ZW

W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW

AU 2000037395 A 20001016 (200106)

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000058466	A2	WO 2000-US6418	20000320
AU 2000037395	A	AU 2000-37395	20000320

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2000037395	A	Based on	WO 200058466

PRIORITY APPLN. INFO: US 2000-178184 20000126; US 1999-126839 19990330; US 1999-140073 19990621; US 1999-147071 19990804; US 1999-160524 19991020; US 1999-160669 19991021; US 1999-172744 19991220

AN 2000-664925 [64] WPIDS

CR 2000-656167 [60]

AB WO 200058466 A UPAB: 20010126

NOVELTY - A **FAS ligand** inhibitory protein (FLINT) analog (I) **resistant** or substantially **resistant** to proteolysis at position 218 of a 271 residue amino acid sequence (S1), fully defined in the specification, in vivo or in vitro, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) (I) which is **resistant** to proteolysis by a serine or threonine **protease**;

(2) (I) comprising an amino acid substitution in the region of position 214-222 of (S1);

(3) (I) comprising an amino acid substitution selected from Arg to Gln, Glu, Ala, Gly, Ser, Val, Tyr or Asn at position 218, Thr to Pro at poisiton 216, and Pro to Tyr at position 217;

(4) (I) comprising an amino acid substitution at position 218 and the substitutions Arg to Asn at position 34, Asp to Thr at position 36, Asp to Asn at position 194, and/or Ser to Thr at position 196;

(5) (I) comprising a Thr to Pro substitution at position 216, and Arg to Gln at position 218;

(6) a pharmaceutical formulation comprising (I);

(7) a fusion protein comprising (I);

(8) a nucleic acid (II) encoding (I);

(9) a formulation adapted for inhibiting ischemic injury during organ transplantation comprising (I) as an active ingredient;

(10) a liquid medium for infusion and preservation of organs comprising (I);

(11) (I) encoded by a nucleic acid that hybridizes under high stringency conditions to an 813 nucleotide sequence (S2), fully defined in the specification;

(12) a vector comprising (II);

(13) a recombinant host cell comprising the vector of (8);

(14) preparation of (I), comprising altering the amino acid sequence in the region at and/or between positions 214 through 222 of (S1); and

(15) a nucleic acid that encodes (I) hybridizing to (S2) under high stringency conditions.

ACTIVITY - Antiinflammatory; antiulcer; vasotropic; immunosuppressive; antiarthritic; antipsoriatic; antithyroid; thyromimetic; neuroprotective; antiviral; cardiant; vulnerary; antiarteriosclerotic; antibacterial; antianemic; hepatotropic; nephrotropic; antirheumatic; nootropic; osteopathic.

MECHANISM OF ACTION - Apoptosis inhibitor; The antiapoptotic activity of (I) was tested in vitro. **FasL** interactions with membrane-bound Fas receptor and LIGHT, inhibitor. The anti-apoptotic activity of (I) was tested in vitro, 25 micro l of Jurkat cells (5X104 cells/well) were added to each well of a 96-well plate and mixed with 25 micro l of recombinant human **FasL** and either 50 micro l of FLINT or FLINT analog. Serial dilutions ranging from 0-1 mg/ml were tested in the assay. Cells were incubated at 37 deg. C overnight. 20 micro l of MTS tetrazolium compound was added to each well and the incubation carried out for 2 hours at 37 deg. C. Absorbance at 490 nm was recorded using a plate reader. Analogs that changed arginine at position 218 to glutamine, glutamic acid, alanine, glycine, or valine showed activity in this assay.

USE - (I) is useful for treating acute lung injury, acute respiratory distress syndrome or ulcerative colitis. (I) is also used for inhibiting T-cell activation, inhibiting ischemic injury during organ transplantation and in liquid mediums for infusion and preservation of organs (claimed). The FLINT analog is used to prevent or treat chronic obstructive pulmonary disease (COPD) and

pulmonary fibrosis (PF). FLINT analogs are used to prevent rejection, in the treatment of autoimmune diseases, and in treating systemic inflammatory responses. FLINT is used for treating analog inflammatory/autoimmune diseases such as rheumatoid arthritis, inflammatory bowl disease, graft-versus-host disease, insulin-dependent diabetes, sepsis, pancreatitis, psoriasis, multiple sclerosis, Hashimoto's throiditis, Grave's disease, infectious diseases such as human immunodeficiency virus (HIV)-induced lymphopenia, fulminant viral hepatitis B/C, chronic hepatitis/cirrhosis, Helicobacter pylori-associated ulceration, ischemia, Reperfusion conditions such as acute coronary syndrome, acute myocardial infarction, congestive heart failure, atherosclerosis, acute cerebral ischemia/infarction, brain/spinal cord trauma, organ preservation during transplant and for treating cytoprotection during cancer treatment, adjuvant to chemotherapy, Alzheimer's, chronic glomerulonephritis, osteoporosis, aplastic anemia, and myelodysplasia. FLINT analogs inhibit the binding of FAS to **FASL** and LIGHT to LT beta R and TR2/HVEM receptors, and can be used to treat or prevent a disease and/or condition that may be associated with the binding. Runaway apoptosis, a condition caused by excessive activation of the FAS/**FASL** signal transduction pathway that can be treated with the FLINT analogs. FLINT analog is useful prophylactically to prevent apoptosis associated with ischemia.

ADVANTAGE - (I) is **resistant** to proteolysis by trypsin-like enzymes and has a half-life which is 2-100 fold greater than FLINT.

Dwg.0/7

L9 ANSWER 4 OF 31 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
ACCESSION NUMBER: 2000-656167 [63] WPIDS
CROSS REFERENCE: 2000-664925 [60]
DOC. NO. CPI: C2000-198584
TITLE: **FAS Ligand** Inhibitory Protein analogs useful for treating abnormal apoptosis related diseases e.g. acute lung injury, pulmonary fibrosis, chronic obstructive pulmonary disease ulcerative colitis or Crohn's disease.
DERWENT CLASS: B04 D16
INVENTOR(S): BECKER, G W; COHEN, F J; GONZALEZ-DEWHITT, P A; HALE, J E; MICANOVIC, R; NEWTON, C M; NOBLITT, T W; RATHMACHALAM, R; TSCHANG, S R; WITCHER, D R; WROBLEWSKI, V J
PATENT ASSIGNEE(S): (ELIL) LILLY & CO ELI
COUNTRY COUNT: 91
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG

WO 2000058465 A2 20001005 (200063)* EN 114
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC
MW NL OA PT SD SE SL SZ TZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CR CU CZ DE DK DM
DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR
KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX NO NZ PL PT RO
RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW
AU 2000037394 A 20001016 (200106)

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000058465 A2		WO 2000-US6417	20000320
AU 2000037394 A		AU 2000-37394	20000320

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000037394 A	Based on	WO 200058465

PRIORITY APPLN. INFO: US 2000-183398 20000218; US 1999-126839 19990330; US 1999-140077 19990621; US 1999-140156 19990621; US 1999-160566 19991020

AN 2000-656167 [63] WPIDS
CR 2000-664925 [60]
AB WO 200058465 A UPAB: 20010126
NOVELTY - A **FAS Ligand** Inhibitory Protein (FLINT) analog comprising a modified form of a 271 residue amino acid sequence (S1), fully defined in the specification, or its fragment, having FLINT biological activity, is new.

DETAILED DESCRIPTION - A **FAS Ligand** Inhibitory Protein (FLINT) analog comprising a modified form of a 271 residue amino acid sequence (S1), fully defined in the specification, or its fragment, having FLINT biological activity, is new. (I) comprises the substitution:

(a) Trp to Asp at position 53, Thr to Pro at position 88, Ala to Ser, Glu or Thr at position 107, Ile to Thr or Glu at position 110 or Pro to Ser at position 104;

(b) Ala to Asn at position 2 or 12, Pro to Asn at position 25, 38, 126 or 171, Arg to Asn at position 35, Ser to Asp at position 37 and Pro to any amino acid at position 38, Ser to Asn at position 166, Leu to Asn at position 172, Asp to Asn at position 194 or Thr to Asn at position 114 and Pro to any amino acid at position 115;

(c) Asn to Trp at position 63, Gly to Asp at position 67 and Ala at position 94 or Gly at position 95 to Tyr, Arg to Glu at

position 69, Arg to Glu or Thr at position 82, Ala to Tyr at position 94 and Gly to Asp at position 95, Phe to Glu at position 96, Ala to Thr at position 101, or Gly to Asp at position 95;

(d) Arg to Glu, Asn, Ser or Thr at position 10, where if the substitution is to Asn then Ala is substituted by Ser or Thr at position 12, Glu to Gln, Asn, Ser or Thr at position 13, where if the substitution is to Asn then Gly is substituted by Ser or Thr at position 15, Glu to Gln, Asn, Ser, or Thr at position 16, where if the substitution is to Asn then Leu is substituted by Ser or Thr at position 18, Arg to Gln, Asn, Ser or Thr at position 17, where if the substitution is to Asn then Val is substituted by Ser or Thr at position 19, Arg to Gln, Asn, Ser or Thr at position 17, where if the substitution is to Asn then Cys is substituted by Ser or Thr at position 33, Arg to Gln, Asn, Ser or Thr at position 34, where if the substitution is to Asn then Asp is substituted by Ser or Thr at position 36, Arg to Gln, Asn, Ser or Thr at position 35, Asp to Gln, Asn, Ser or Thr at position 36, where if the substitution is to Asn then Pro is substituted by Ser or Thr at position 38, Arg to Gln, Asn, Ser or Thr at position 143, where if the substitution is to Asn then Cys is substituted by Ser or Thr at position 145, or Asp to Gln, Asn, Ser or Thr at position 161, where if the substitution is to Asn then Leu is substituted by Ser or Thr at position 163;

(e) Ala to Thr position 2, 12,107, 179 or 209, Thr to Ala at position 4 or 162, Val at position 1, or Ile at position 110 to Met; Glu to Asp at position 13, Arg to Trp at position 17, Ala to Pro at position 75, Ser to Leu at position 102, Gly to Ala at position 169, Glu to Lys at position 183, Gln to Arg at position 225, Gly to Glu at position 237, or Val to Gly at position 270; or

(f) Ala to Asn at position 12 and Glu to Gln at position 13, Arg to Asn at position 34 and Asp to Thr at position 36, Arg to Asp at position 35 and Ser to Thr at position 37, Ser to Asn at position 132 and Ser to Thr at position 134, Asp to Asn at position 194 and Ser to Thr at position 196, Arg at position 35 and Asp at position 194 to Asn, Ala to Asn at position 12 Glu to Gln at position 13 Asp to Asn at position 194 and Ser to Thr at position 194, Arg to Asp at position 34 Asp to Asn at position 194 and Ser to Thr at position 196, Arg at position 35 and Asp at position 194 to Asn and Ser to Thr at position 37 and/or 196, or Arg to Gln at position 218.

INDEPENDENT CLAIMS are also included for the following:

(1) a polypeptide fragment (II) of FLINT which is biologically active in vivo or in vitro;

(2) a fusion protein having a formula (III) ;

(3) a nucleic acid (IV) encoding (I), (II) or the fusion protein;

(4) a polynucleotide (V) comprising (IV);

(5) a vector (VI) comprising (IV) or (V);

(6) a host cell transformed with (VI); and

(7) a pharmaceutical composition comprising (I).

Fc = Fc fragment of an antibody; and

X = independently, a peptide derivative of (I) or (II) or **protease resistant** FLINT analog.

X is covalently linked at its C terminus to the N terminus of Fc fragment of the antibody.

ACTIVITY - Antiinflammatory; antiapoptopic; antiulcer; cytostatic. BALB/c mice were given intravenous injections in the lateral tail vein of 6 mg of D(+)- Galactosamine and 100 ml of phosphate buffer saline in 3 micro g of lipopolysaccharide (LPS) B Escherichia coli in 100 micro l of phosphate buffered saline (PBS). After LPS challenge, the animals were injected intraperitonially with (I) (200 micro g). Suitable controls included hamster immunoglobulin (Ig)G (500 micro g), monoclonal antibody (mAb) against murine tumor necrosis factor, TN3-19.12 and anti-mouse **Fas Ligand** at 0, 2, 4, 6 hour-point respectively.
The survival rates of the mice were determined 24 and 48 hours after LPS injection. The results showed that FLINT analog was effective in protecting animals from acute liver damage.

MECHANISM OF ACTION - Inhibitor of binding of **FasL** to Fas or LIGHT to LT beta R and/or TR2/herpes virus entry mediator.

USE - (I) is useful for treating a patient suffering from disease or condition relating to abnormal apoptosis such as acute lung injury, acute respiratory distress syndrome, pulmonary fibrosis, chronic obstructive pulmonary disease, ulcerative colitis, or Crohn's disease.
Dwg.0/2

L9 ANSWER 5 OF 31 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2000435663 MEDLINE
DOCUMENT NUMBER: 20359909 PubMed ID: 10683315
TITLE: CpG-specific common commitment in caspase-dependent and -independent cell deaths.
AUTHOR: Qi L; Sit K H
CORPORATE SOURCE: Department of Anatomy, Faculty of Medicine, National University of Singapore, Kent Ridge, 119260, Singapore.
SOURCE: MOLECULAR CELL BIOLOGY RESEARCH COMMUNICATIONS, (2000 Jan) 3 (1) 33-41.
Journal code: DRR; 100889076. ISSN: 1522-4724.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200009
ED Entered STN: 20000928
Last Updated on STN: 20000928
Entered Medline: 20000915
AB Cell death in mammals seems to have caspase-dependent and

-independent pathways unlike that in Caenorhabditis elegans where CED-3 **pr tease** activation is the central command. A recent suggestion to define apoptosis as the caspase-dependent or caspase-committed cell death form and leave cell death committed by other pathways as just cell death was meant to categorize the apparent divergence in mammalian cell death pathways. However, we show CpG oligonucleotides (ODN) blocking caspase-dependent fas(CD95) ligand-mediated apoptosis as well as caspase-independent etoposide-mediated apoptosis and etoposide-zVAD-mediated necrosis. CpG specificity was demonstrated by reversing the CpG motif or replacing it with a methylated motif (mCpG) which failed to inhibit. CpG ODN blocked CpG-specific DNA cleavage by rare-cutting NotI restriction, which produced a megabase cleavage pattern similar to that in the **fasL** and etoposide cell death inductions. CpG ODN inhibition was similar to that by CpG-specific SssI methylase. A common CpG-specific commitment point preceding caspase-dependent and -independent cell death pathways was suggested. CpG-specific modulation is a key epigenetic mechanism in genomic imprinting, **resisting** nuclease restriction, and patterning of chromatin conformations. It is now shown to have a powerful effect modulating cell death.

L9 ANSWER 6 OF 31 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD

ACCESSION NUMBER: 1999-229531 [19] WPIDS
DOC. NO. CPI: C1999-067564
TITLE: **Protease-resistant Fas ligand** derivatives used for prevention of, e.g. cancer.
DERWENT CLASS: B04 D16
INVENTOR(S): NAGATA, S; TANAKA, M
PATENT ASSIGNEE(S): (MOCH) MOCHIDA PHARM CO LTD; (OSAB-N) OSAKA BIOSCIENCE INST
COUNTRY COUNT: 22
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9914325	A1	19990325	(199919)*	JA	60
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE					
W: CA JP US					
EP 1016721	A1	20000705	(200035)	EN	
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE					

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE

WO 9914325	A1	WO 1998-JP4187	19980917
EP 1016721	A1	EP 1998-943025	19980917
		WO 1998-JP4187	19980917

FILING DETAILS:

PATENT NO	KIND	PATENT NO
EP 1016721	A1 Based on	WO 9914325

PRIORITY APPLN. INFO: JP 1997-252541 19970917
AN 1999-229531 [19] WPIDS
AB WO 9914325 A UPAB: 19990518
NOVELTY - **Protease-resistant Fas ligand** derivatives in which a region of human **Fas ligand** is susceptible to **protease** attack has been deleted, are new.

DETAILED DESCRIPTION - Derivatives of human **Fas ligand** (and DNA encoding them) in which amino acid residues 129 (lysine) and 130 (glutamine) have been deleted or substituted by other amino acids, and in which at least one of the residues 111-128 and 131-333 has similarly been deleted or substituted, are new.

An INDEPENDENT CLAIM is also included for apoptosis modulators containing soluble **Fas ligand**.

ACTIVITY - The modification in the **Fas ligand** renders it **resistant** to the action of **proteases** such as the metalloproteinase which in vivo cleaves the active membrane-bound **Fas ligand** (which is active as an apoptosis inducer).

USE - For the prevention and treatment of diseases such as cancer, viral infection and autoimmune disease, e.g. by introduction of DNA encoding the modified **Fas ligand** into effector cells using a suitable gene therapy vector.

Dwg.0/7

L9 ANSWER 7 OF 31 MEDLINE
ACCESSION NUMBER: 1999233669 MEDLINE
DOCUMENT NUMBER: 99233669 PubMed ID: 10216102
TITLE: Anticancer drugs induce caspase-8/FLICE activation and apoptosis in the absence of CD95 receptor/ligand interaction.
AUTHOR: Wesselborg S; Engels I H; Rossmann E; Los M; Schulze-Osthoff K
CORPORATE SOURCE: Department of Internal Medicine I, Eberhard-Karls-University, Tubingen, Germany.
SOURCE: BLOOD, (1999 May 1) 93 (9) 3053-63.
Journal code: A8G; 7603509. ISSN: 0006-4971.
PUB. COUNTRY: United States

Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199905
ED Entered STN: 19990601
Last Updated on STN: 20000303
Entered Medline: 19990518

AB **Proteases** of the caspase family are the critical executioners of apoptosis. Their activation has been mainly studied upon triggering of death receptors, such as CD95 (Fas/APO-1) and tumor necrosis factor-R1, which recruit caspase-8/FLICE as the most proximal effector to the receptor complex. Because apoptosis induced by anticancer drugs has been proposed to involve CD95/CD95 ligand interaction, we investigated the mechanism of caspase activation by daunorubicin, doxorubicin, etoposide, and mitomycin C. In Jurkat leukemic T cells, all drugs induced apoptosis and the cleavage of procaspase-8 to its active p18 subunit. However, cells **resistant** to CD95 were equally susceptible to anticancer drugs and activated caspase-8 with a similar kinetic and dose response as CD95-sensitive cells. The broad caspase inhibitor benzyloxycarbonyl-Val-Ala-Asp-fluoromethylketone prevented apoptosis and caspase-8 activation in response to CD95 and drug treatment, whereas a neutralizing CD95 decoy as well as a dominant-negative FADD construct selectively abrogated CD95, but not drug-induced effects. A potent activation of caspase-8 was also induced by cycloheximide, indicating that it was independent of protein synthesis. Our data, therefore, show that (1) anticancer drug-induced apoptosis does not require de novo synthesis of death ligands or CD95 interaction, and (2) that caspase-8 can be activated in the absence of a death receptor signaling.

L9 ANSWER 8 OF 31 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 2000050586 MEDLINE
DOCUMENT NUMBER: 20050586 PubMed ID: 10583368
TITLE: alpha-fetoprotein causes apoptosis in tumor cells via a pathway independent of CD95, TNFR1 and TNFR2 through activation of caspase-3-like proteases.
AUTHOR: Dudich E; Semenkova L; Dudich I; Gorbatova E; Tochtamisheva N; Tatulov E; Nikolaeva M; Sukhikh G
CORPORATE SOURCE: Institute of Engineering Immunology, Lyubuchany, Moscow Region, Russia.. dudich@ineos.ac.ru
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1999 Dec) 266 (3) 750-61.
Journal code: EMZ; 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

ENTRY MONTH: 200001
ED Entered STN: 20000209
Last Updated on STN: 20000525
Entered Medline: 20000131

AB alpha-Fetoprotein (AFP) is an oncoembryonal protein with multiple cell growth regulating, differentiating and immunosuppressive activities. Previous studies have shown that treatment of tumor cells in vitro with 1-10 microM AFP produces significant suppression of tumor cell growth by inducing dose-dependent cytotoxicity, but the molecular mechanisms underlying these AFP functions are obscure. Here, we show that AFP cytotoxicity is closely related to apoptosis, as shown by cell morphology, nuclear DNA fragmentation and caspase-3-like activity resulting in cleavage of poly(ADP-ribose) polymerase. Apoptosis was significantly inhibited by a CPP32 family **protease** inhibitor whereas a general caspase inhibitor had no inhibitory effect, showing some enhancement of AFP-mediated cell death. Using fluorogenic caspase substrates, we found that caspase-3-like **proteases** were activated as early as 4 h after treatment of Raji cells with 15 microM AFP, whereas caspase-1, caspase-8, and caspase-9-like activity was not detected during the time interval 0.5-17 h. AFP treatment of Raji cells increased Bcl-2 protein, showing that AFP-induced apoptosis is not explained by downregulation of the Bcl-2 gene. This also suggests that AFP operates downstream of the Bcl-2-sensitive step. AFP notably decreased basal levels of soluble and membrane-bound **Fas ligand**. Incubation of AFP-sensitive tumor cells (HepG2, Raji) with neutralizing anti-Fas, anti-tumor necrosis factor receptor (TNFR)1 or anti-TNFR2 mAb did not prevent AFP-induced apoptosis, demonstrating its independence of Fas-dependent and TNFR-dependent signaling. In addition, it was found that cells **resistant** to TNF-induced (Raji) or Fas-induced (MCF-7) apoptosis are, nevertheless, sensitive to AFP-mediated cell death. In contrast, cells sensitive to Fas-mediated cell death (Jurkat) are completely **resistant** to AFP. Taken as a whole, our data demonstrate that: (a) AFP induces apoptosis in tumor cells independently of Fas/**Fas ligand** or TNFR/TNF signaling pathways, and (b) AFP-mediated cell death involves activation of the effector caspase-3-like **proteases**, but is independent of upstream activation of the initiator caspase-1, caspase-8, and caspase-9-like **proteases**.

L9 ANSWER 9 OF 31 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 1999413939 MEDLINE
DOCUMENT NUMBER: 99413939 PubMed ID: 10484385
TITLE: Cryptosporidium parvum induces apoptosis in biliary epithelia by a Fas/**Fas ligand** -dependent mechanism.
AUTHOR: Chen X M; Gores G J; Paya C V; LaRusso N F

CORPORATE SOURCE: Center for Basic Research in Digestive Diseases, Division of Gastroenterology and Hepatology, Mayo Medical School, Clinic and Foundation, Rochester, Minnesota 55905, USA.
CONTRACT NUMBER: DK-24031 (NIDDK)
SOURCE: AMERICAN JOURNAL OF PHYSIOLOGY, (1999 Sep) 277 (3 Pt 1) G599-608.
Journal code: 3U8; 0370511. ISSN: 0002-9513.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199910
ED Entered STN: 20000111
Last Updated on STN: 20000111
Entered Medline: 19991028

AB Although the clinical features of sclerosing cholangitis from opportunistic infections of the biliary tree in patients with acquired immunodeficiency syndrome (AIDS) are well known, the mechanisms by which associated pathogens, such as Cryptosporidium parvum, cause disease are obscure. Using an in vitro model of biliary cryptosporidiosis, we observed that C. parvum induces apoptosis in cultured human biliary epithelia. Both caspase **protease** inhibitors and neutralizing antibodies to either Fas receptor (Fas) and **Fas ligand** (**FasL**) inhibited this process; neutralizing antibodies to other apoptotic cytokines [interleukin-1beta (IL-1beta), tumor necrosis factor-alpha (TNF-alpha), and transforming growth factor-beta (TGF-beta)] had no effect. C. parvum stimulated **FasL** membrane surface translocation, increased both **FasL** and Fas protein expression in infected biliary epithelia, and induced a marked increase of soluble **FasL** (but not IL-1beta, TNF-alpha, and TGF-beta) in supernatants from infected cells. When a coculture model is used in which infected and uninfected cell populations were physically separated by a semipermeable membrane, both uninfected biliary epithelia and uninfected Fas-sensitive Jurkat cells (but not a Fas-**resistant** Jurkat cell line) underwent apoptosis when cocultured with infected biliary epithelia. Moreover, both a neutralizing antibody to **FasL** and a metalloprotease inhibitor blocked the apoptosis in uninfected cocultured cells. Activation of caspase activity was also observed in uninfected cocultured biliary epithelia. The data suggest that C. parvum induces apoptosis in biliary epithelia by a Fas/**FasL** -dependent mechanism involving both autocrine and paracrine pathways. These observations may be relevant to both the pathogenesis and therapy of the cholangitis seen in AIDS patients with biliary cryptosporidiosis.

L9 ANSWER 10 OF 31 SCISEARCH COPYRIGHT 2001 ISI (R)
ACCESSION NUMBER: 1999:691935 SCISEARCH
THE GENUINE ARTICLE: 233BF
TITLE: Cryptosporidium parvum induces apoptosis in biliary epithelia by a Fas/**Fas ligand** -dependent mechanism
AUTHOR: Chen X M; Gores G J; Paya C V; LaRusso N F (Reprint)
CORPORATE SOURCE: MAYO CLIN & MAYO FDN, CTR BASIC RES DIGEST DIS, DIV GASTROENTEROL & HEPATOL, 200 1ST ST SW, ROCHESTER, MN 55905 (Reprint); MAYO CLIN & MAYO FDN, CTR BASIC RES DIGEST DIS, DIV GASTROENTEROL & HEPATOL, ROCHESTER, MN 55905; MAYO CLIN & MAYO FDN, DIV EXPT PATHOL, ROCHESTER, MN 55905
COUNTRY OF AUTHOR: USA
SOURCE: AMERICAN JOURNAL OF PHYSIOLOGY-GASTROINTESTINAL AND LIVER PHYSIOLOGY, (SEP 1999) Vol. 40, No. 3, pp. G599-G608.
Publisher: AMER PHYSIOLOGICAL SOC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814.
ISSN: 0193-1857.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 61

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Although the clinical features of sclerosing cholangitis from opportunistic infections of the biliary tree in patients with acquired immunodeficiency syndrome (AIDS) are well known, the mechanisms by which associated pathogens, such as Cryptosporidium parvum, cause disease are obscure. Using an in vitro model of biliary cryptosporidiosis, we observed that C. parvum induces apoptosis in cultured human biliary epithelia. Both caspase **protease** inhibitors and neutralizing antibodies to either Fas receptor (Fas) and **Fas ligand** (**FasL**) inhibited this process; neutralizing antibodies to other apoptotic cytokines [interleukin-1 beta (IL-1 beta), tumor necrosis factor-alpha (TNF-alpha), and transforming growth factor-p (TGF-P)I had no effect. C, parvum stimulated Fast membrane surface translocation, increased both Fast and Fas protein expression in infected biliary epithelia, and induced a marked increase of soluble Fast (but not IL-1 beta, TNF-alpha, and TGF-beta) in supernatants from infected cells. When a coculture model is used in which infected and uninfected cell populations were physically separated by a semipermeable membrane, both uninfected biliary epithelia and uninfected Fas-sensitive Jurkat cells (but not a Fas-**resistant** Jurkat cell line) underwent apoptosis when cocultured with infected biliary epithelia. Moreover, both a neutralizing antibody to Fast and a metalloprotease inhibitor

blocked the apoptosis in uninfected cocultured cells. Activation of caspase activity was also observed in uninfected cocultured biliary epithelia. The data suggest that C. parvum induces apoptosis in biliary epithelia by a Fas/**FasL**-dependent mechanism involving both autocrine and paracrine pathways. These observations may be relevant to both the pathogenesis and therapy of the cholangitis seen in AIDS patients with biliary cryptosporidiosis.

L9 ANSWER 11 OF 31 MEDLINE DUPLICATE 5
ACCESSION NUMBER: 1999102257 MEDLINE
DOCUMENT NUMBER: 99102257 PubMed ID: 9884343
TITLE: Toxic bile salts induce rodent hepatocyte apoptosis via direct activation of Fas.
AUTHOR: Faubion W A; Guicciardi M E; Miyoshi H; Bronk S F; Roberts P J; Svingen P A; Kaufmann S H; Gores G J
CORPORATE SOURCE: Division of Gastroenterology and Hepatology, Mayo Medical School, Clinic, and Foundation, Rochester, Minnesota 55905, USA.
CONTRACT NUMBER: CA-69008 (NCI)
DK-41876 (NIDDK)
SOURCE: JOURNAL OF CLINICAL INVESTIGATION, (1999 Jan) 103 (1) 137-45.
Journal code: HS7; 7802877. ISSN: 0021-9738.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199902
ED Entered STN: 19990301
Last Updated on STN: 20000303
Entered Medline: 19990216

AB Cholestatic liver injury appears to result from the induction of hepatocyte apoptosis by toxic bile salts such as glycochenodeoxycholate (GCDC). Previous studies from this laboratory indicate that cathepsin B is a downstream effector **protease** during the hepatocyte apoptotic process. Because caspases can initiate apoptosis, the present studies were undertaken to determine the role of caspases in cathepsin B activation. Immunoblotting of GCDC-treated McNtcp.24 hepatoma cells demonstrated cleavage of poly(ADP-ribose) polymerase and lamin B1 to fragments that indicate activation of effector caspases. Transfection with CrmA, an inhibitor of caspase 8, prevented GCDC-induced cathepsin B activation and apoptosis. Consistent with these results, an increase in caspase 8-like activity was observed in GCDC-treated cells. Examination of the mechanism of GCDC-induced caspase 8 activation revealed that dominant-negative FADD inhibited apoptosis and that hepatocytes isolated from Fas-deficient lymphoproliferative mice were **resistant** to GCDC-induced apoptosis. After GCDC

treatment, immunoprecipitation experiments demonstrated Fas oligomerization, and confocal microscopy demonstrated DeltaFADD-GFP (Fas-associated death domain-green fluorescent protein, aggregation in the absence of detectable **Fas ligand** mRNA. Collectively, these data suggest that GCDC-induced hepatocyte apoptosis involves ligand-independent oligomerization of Fas, recruitment of FADD, activation of caspase 8, and subsequent activation of effector **proteases**, including downstream caspases and cathepsin B.

L9 ANSWER 12 OF 31 MEDLINE
ACCESSION NUMBER: 1998261459 MEDLINE
DOCUMENT NUMBER: 98261459 PubMed ID: 9596680
TITLE: Caspase activation is required for nitric oxide-mediated, CD95(APO-1/Fas)-dependent and independent apoptosis in human neoplastic lymphoid cells.
AUTHOR: Chlichlia K; Peter M E; Rocha M; Scaffidi C; Bucur M; Krammer P H; Schirrmacher V; Umansky V
CORPORATE SOURCE: Division of Cellular Immunology and the Division of Immunogenetics, Tumor Immunology Program, German Cancer Research Center, Heidelberg, Germany.
SOURCE: BLOOD, (1998 Jun 1) 91 (11) 4311-20.
Journal code: A8G; 7603509. ISSN: 0006-4971.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199806
ED Entered STN: 19980713
Last Updated on STN: 20000303
Entered Medline: 19980626

AB Nitric oxide (NO), an important effector molecule involved in immune regulation and host defense, was shown to induce apoptosis in lymphoma cells. In the present report the NO donor glycerol trinitrate was found to induce apoptosis in Jurkat cells that are sensitive to CD95-mediated kill. In contrast, a CD95-**resistant** Jurkat subclone showed substantial protection from apoptosis after exposure to NO. NO induced mRNA expression of CD95 (APO-1/Fas) and TRAIL/APO-2 ligands. Moreover, NO triggered apoptosis in freshly isolated human leukemic lymphocytes which were also sensitive to anti-CD95 treatment. The ability of NO to induce apoptosis was completely blocked by a broad-spectrum ICE (interleukin-1beta converting enzyme)-**protease**/caspase inhibitor and correlated with FLICE/caspase-8 activation. This activation was abrogated in some neoplastic lymphoid cells but not in others by the inhibitor of protein synthesis cycloheximide. Our results were confirmed using an in vitro experimental model of

coculture of human lymphoid target cells with activated bovine endothelial cells generating NO as effectors. Furthermore, the inhibition of endogenous NO production with the inducible NO synthase inhibitor NG-monomethyl-L-arginine caused a complete abrogation of the apoptotic effect. Our data provide evidence that NO-induced apoptosis in human neoplastic lymphoid cells strictly requires activation of caspases, in particular FLICE, the most CD95 receptor-proximal caspase. Depending on the cell line tested this activation required or was independent of the CD95 receptor/ligand system.

L9 ANSWER 13 OF 31 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 1998217202 MEDLINE
DOCUMENT NUMBER: 98217202 PubMed ID: 9558119
TITLE: Disparate roles for TNF-alpha and **Fas ligand** in concanavalin A-induced hepatitis.
AUTHOR: Ksontini R; Colagiovanni D B; Josephs M D; Edwards C K 3rd; Tannahill C L; Solorzano C C; Norman J; Denham W; Clare-Salzler M; MacKay S L; Moldawer L L
CORPORATE SOURCE: Department of Surgery, University of Florida College of Medicine, Gainesville 32610, USA.
CONTRACT NUMBER: GM-40586 (NIGMS)
SOURCE: JOURNAL OF IMMUNOLOGY, (1998 Apr 15) 160 (8) 4082-9. Journal code: IFB. ISSN: 0022-1767.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199805
ED Entered STN: 19980514
Last Updated on STN: 20000303
Entered Medline: 19980504

AB Apoptosis is a physiologic process that serves to eliminate cells during development or in response to immunologic regulation. In acute inflammation, however, apoptosis triggered by the overproduction of "death factors" such as TNF-alpha or **Fas ligand** (**FasL**) may contribute to tissue injury. Both TNF-alpha and **FasL** are presumed to convey an apoptotic signal by activating a cascade of cysteine-aspartate **proteases**, which includes IL-1beta-converting enzyme or caspase-1. In the present study, we evaluated the contribution of TNF-alpha and **FasL**, as well as the role of caspase-1, in Con A-induced hepatitis. We report here that TNF-alpha and **FasL** mRNA and protein levels are both increased in the livers of Con A-challenged mice. Using a novel inhibitor of TNF-alpha, we can confirm that Con A-induced hepatitis is primarily TNF-alpha dependent. Blockade of **FasL** with a soluble Fas immunoadhesin does not prevent liver injury in animals treated with

Con A alone. However, administration of a matrix metalloproteinase inhibitor exacerbates liver injury, in part through a **FasL** -dependent process, since pretreatment with the soluble Fas immunoadhesin reduces liver injury in this model. In addition, mice lacking functional caspase-1 are **resistant** to Con A-induced hepatitis, even after pretreatment with a matrix metalloproteinase inhibitor. We conclude that TNF-alpha plays a predominant role in Con A-induced liver injury, although concomitant activation of **FasL** can also lead to apoptotic injury. Furthermore, Con A-induced hepatitis is caspase-1 dependent.

L9 ANSWER 14 OF 31 MEDLINE DUPLICATE 7
ACCESSION NUMBER: 1999075976 MEDLINE
DOCUMENT NUMBER: 99075976 PubMed ID: 9858879
TITLE: Anti-Fas antibody differentially regulates apoptosis in **Fas ligand resistant** carcinoma lines via the caspase 3 family of cell death **proteases** but independently of bcl2 expression.
AUTHOR: Crowe D L; Boardman M L; Fong K S
CORPORATE SOURCE: Center for Craniofacial Molecular Biology, University of Southern California, Los Angeles 90033, USA.
CONTRACT NUMBER: DE07211 (NIDCR)
DE10966 (NIDCR)
SOURCE: ANTICANCER RESEARCH, (1998 Sep-Oct) 18 (5A) 3163-70. Journal code: 59L; 8102988. ISSN: 0250-7005.
PUB. COUNTRY: Greece
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199812
ED Entered STN: 19990115
Last Updated on STN: 20000303
Entered Medline: 19981229

AB Deregulation of cell death pathways is an important feature of tumorigenesis. Fas, a member of the tumor necrosis factor receptor superfamily, is a transmembrane protein that can transduce cell death signals via a proteolytic cascade upon crosslinking or ligand binding. Fas has been implicated in the cell turnover of normal stratified squamous epithelia. To determine if altered Fas mediated cell death pathways participate in epithelial tumorigenesis, we examined squamous cell carcinoma (SCC) lines for sensitivity to **Fas ligand** (**FasL**) or an agonistic anti-Fas antibody. All cell lines examined were **resistant** to **FasL** mediated cell death. The carcinoma cell line SCC71 was also highly **resistant** to anti-Fas antibody. Another line, SCC9, underwent rapid cell death with characteristic features of apoptosis after exposure to anti-Fas antibody. However, binding

of both **FasL** and anti-Fas antibody recruited downstream effector molecules to the Fas cytoplasmic domain in both SCC9 and SCC71 cells. Inhibition of the caspase 3- but not the ICE family of cell death **proteases** blocked apoptosis in SCC9 cells independently of expression of the anti-apoptotic protein bcl2. We concluded that Fas differentially mediates apoptosis in SCC lines by activation of caspase 3 family members but independent of bcl2 expression.

L9 ANSWER 15 OF 31 MEDLINE DUPLICATE 8
ACCESSION NUMBER: 1998414272 MEDLINE
DOCUMENT NUMBER: 98414272 PubMed ID: 9743340
TITLE: Caspase dependence of target cell damage induced by cytotoxic lymphocytes.
AUTHOR: Sarin A; Haddad E K; Henkart P A
CORPORATE SOURCE: Experimental Immunology Branch, National Cancer Institute, Bethesda, MD 20892-1360, USA.
SOURCE: JOURNAL OF IMMUNOLOGY, (1998 Sep 15) 161 (6) 2810-6. Journal code: IFB; 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199810
ED Entered STN: 19981020
Last Updated on STN: 20000303
Entered Medline: 19981006

AB Since the CTL secreted granule **protease** granzyme B can activate multiple target caspases, it has been proposed that this pathway is responsible for CTL-induced cytolysis of Fas-negative targets. However, target lysis via the granule exocytosis pathway is completely **resistant** to caspase inhibitors. To test the possibility that granzymes trigger a postcaspase cytoplasmic apoptotic pathway leading to lysis, we have examined the caspase dependence of several cytoplasmic changes associated with apoptotic death. Rapid prelytic phosphatidylserine externalization was induced in Jurkat target cells by both the **Fas ligand** (**FasL**)/Fas and the granule exocytosis effector pathways. This was specifically blocked by peptide ketone caspase inhibitors when induced by the former, but not by the latter, pathway. A rapid prelytic loss of target mitochondrial psi was also induced by both CTL effector pathways, and this was also specifically blocked by caspase inhibitors when induced by the **FasL**/Fas, but not by the granule exocytosis, pathway. Similarly, target membrane blebbing induced by CTL via the **FasL**/Fas, but not via the granule exocytosis, effector pathway was specifically blocked by caspase inhibitors. In contrast to the above nonnuclear damage, CTL-induced target staining by the lipid probe FM1-43 reflecting

plasma membrane endocytosis was blocked by caspase inhibitors. Thus, when caspase activation is blocked, the granule exocytosis pathway triggers several parameters of target apoptotic damage in addition to lysis, suggesting that granzymes directly trigger a postcaspase cytoplasmic apoptotic death pathway.

L9 ANSWER 16 OF 31 MEDLINE
ACCESSION NUMBER: 1998158504 MEDLINE
DOCUMENT NUMBER: 98158504 PubMed ID: 9498739
TITLE: Apoptosis induced by a chimeric Fas/FLICE receptor: lack of requirement for Fas- or FADD-binding proteins.
AUTHOR: Memon S A; Hou J; Moreno M B; Zacharchuk C M
CORPORATE SOURCE: Laboratory of Immune Cell Biology, Division of Basic Sciences, National Cancer Institute, Bethesda, MD 20892, USA.
SOURCE: JOURNAL OF IMMUNOLOGY, (1998 Mar 1) 160 (5) 2046-9. Journal code: IFB; 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199803
ED Entered STN: 19980326
Last Updated on STN: 20000303
Entered Medline: 19980318

AB Current models for Fas (CD95)-mediated apoptosis suggest that FLICE/caspase-8 is recruited and activated, which results in cell death. However, the role of additional molecules in Fas signaling and FLICE activation is not clear. A chimeric Fas/FLICE (F/F) receptor, containing the extracellular/transmembrane portion of Fas and the caspase region of FLICE, mediated anti-Fas apoptosis. FLICE **protease** subunits were generated from the F/F precursor. Killing induced by Fas, but not F/F, was blocked by a dominant negative FADD. Apoptosis triggered through Fas and F/F was inhibited by coexpression of CrmA and p35, but not Bcl-xL. F/F bypassed Fas **resistance** in COS-7 cells and blocking by the death effector domain (DED)-containing viral protein MC159. These results show that: 1) F/F induces cell death, indicating that FLICE activation is sufficient for apoptosis and does not require additional Fas- or FADD-binding proteins; and 2) F/F bypasses proximal defects in Fas signaling that prevent FLICE recruitment or activation.

L9 ANSWER 17 OF 31 MEDLINE DUPLICATE 9
ACCESSION NUMBER: 1998421154 MEDLINE
DOCUMENT NUMBER: 98421154 PubMed ID: 9740801
TITLE: Essential requirement for caspase-8/FLICE in the initiation of the Fas-induced apoptotic cascade.

AUTHOR: Juo P; Kuo C J; Yuan J; Blenis J
CORPORATE SOURCE: Department of Cell Biology, Harvard Medical School, Boston, Massachusetts 02115, USA.
SOURCE: CURRENT BIOLOGY, (1998 Sep 10) 8 (18) 1001-8.
Journal code: B44; 9107782. ISSN: 0960-9822.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199812
ED Entered STN: 19990115
Last Updated on STN: 19990115
Entered Medline: 19981207

AB BACKGROUND: Fas (APO-1/CD95) is a member of the tumor necrosis factor receptor (TNF-R) family and induces apoptosis when crosslinked with either **Fas ligand** or agonistic antibody (Fas antibody). The Fas-**Fas ligand** system has an important role in the immune system where it is involved in the downregulation of immune responses and the deletion of peripheral autoreactive T lymphocytes. The intracellular domain of Fas interacts with several proteins including FADD (MORT-1), DAXX, RIP, FAF-1, FAP-1 and Sentrin. The adaptor protein FADD can, in turn, interact with the cysteine **protease** caspase-8 (FLICE/MACH/Mch5). RESULTS: In a genetic screen for essential components of the Fas-mediated apoptotic cascade, we isolated a Jurkat T lymphocyte cell line deficient in caspase-8 that was completely **resistant** to Fas-induced apoptosis. Complementation of this cell line with wild-type caspase-8 restored Fas-mediated apoptosis. Fas activation of multiple caspases and of the stress kinase p38 and c-Jun NH2-terminal kinase (JNK) was completely blocked in the caspase-8-deficient cell line. Furthermore, the cell line was severely deficient in cell death induced by TNF-alpha and was partially deficient in cell death induced by ultraviolet irradiation, adriamycin and etoposide. CONCLUSIONS: This study provides the first genetic evidence that caspase-8 occupies an essential and apical position in the Fas signaling pathway and suggests that caspase-8 may participate broadly in multiple apoptotic pathways.

L9 ANSWER 18 OF 31 MEDLINE DUPLICATE 10
ACCESSION NUMBER: 1998202673 MEDLINE
DOCUMENT NUMBER: 98202673 PubMed ID: 9541592
TITLE: Differential regulation of TRAIL and CD95 ligand in transformed cells of the T and B lymphocyte lineage.
AUTHOR: Mariani S M; Krammer P H
CORPORATE SOURCE: Division of Immunogenetics, German Cancer Research Center, Heidelberg, Germany.
SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (1998 Mar) 28 (3)

973-82.
Journal code: EN5; 1273201. ISSN: 0014-2980.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199804
ED Entered STN: 19980430
Last Updated on STN: 20000303
Entered Medline: 19980423

AB TRAIL (APO-2 ligand) and CD95L (CD95/APO-1/**Fas ligand**) share the highest homology among the TNF family members and the ability to induce apoptosis. These similarities raise the issue of a potential functional redundancy between the two ligands. We have previously shown that CD95L-**resistant** cells may be sensitive to TRAIL, even though apoptosis induced by both ligands is blocked by caspase inhibitors. Here we investigated TRAIL protein expression in cells of T and B origin and compared its regulation of expression with that of CD95L. A rabbit antibody (Ab) to a peptide sequence in the extracellular region of TRAIL identified recombinant TRAIL (rTRAIL) produced by Sf9 cells as a protein of approximately 32-33 kDa and soluble rTRAIL as a 19-20-kDa protein. In human and mouse cells, the Ab identified a 33-34-kDa and an additional 19-20-kDa protein only in human cells. Both transformed cells of the T and B lymphocyte lineage were found to react with the anti-TRAIL Ab by immunoblot analysis and surface staining. The majority of the cells analyzed co-expressed TRAIL and CD95L. Two cell lines showed a mirror-pattern, one being TRAILhigh CD95Llow and the other TRAILlow CD95Lhigh, thus suggesting the existence of a cell type-specific regulation of expression of the two ligands. Differently from CD95L, surface TRAIL was not up-regulated by any of the metalloprotease inhibitors tested, independently of the cell type analyzed. Conversely, reactivity with the anti-TRAIL but not with the anti-CD95L Ab was enhanced by cysteine **protease** inhibitors. An in vitro cleavage assay showed that generation of soluble rTRAIL was dependent on the functional activity of cysteine **proteases**, as it was blocked by leupeptin and E64 but not by the metalloprotease inhibitor 1,10-phenanthroline. Thus, even though TRAIL and CD95L share structural and functional properties, they have unique properties as they differ in their regulatory pathways, i.e. cell-type-dependent expression and sensitivity to **protease** inhibitors.

L9 ANSWER 19 OF 31 MEDLINE
ACCESSION NUMBER: 1998437162 MEDLINE
DOCUMENT NUMBER: 98437162 PubMed ID: 9761753
TITLE: Oxygen toxicity in mouse lung: pathways to cell

death.

AUTHOR: Barazzone C; Horowitz S; Donati Y R; Rodriguez I; Piguet P F

CORPORATE SOURCE: Departments of Pediatrics and Pathology, University of Geneva, Switzerland.. Constance.Barazzone@medecine.unige.ch

SOURCE: AMERICAN JOURNAL OF RESPIRATORY CELL AND MOLECULAR BIOLOGY, (1998 Oct) 19 (4) 573-81. Journal code: AOB; 8917225. ISSN: 1044-1549.

PUB. COUNTRY: United States

Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199811

ED Entered STN: 19990106
Last Updated on STN: 19990106
Entered Medline: 19981105

AB Mice exposed to 100% O2 die after 3 or 4 d with diffuse alveolar damage and alveolar edema. Extensive cell death is evident by electron microscopy in the alveolar septa, affecting both endothelial and epithelial cells. The damaged cells show features of both apoptosis (condensation and margination of chromatin) and necrosis (disruption of the plasma membrane). The electrophoretic pattern of lung DNA indicates both internucleosomal fragmentation, characteristic of apoptosis, and overall degradation, characteristic of necrosis. Hyperoxia induces a marked increase in RNA or protein levels of p53, bax, bcl-x, and Fas, which are known to be expressed in certain types of apoptosis. However, we did not detect an increased activity of **proteases** belonging to the apoptosis "executioner" machinery, such as CPP32 (caspase 3), ICE (caspase 1), or cathepsin D. Furthermore, administration of an ICE-like **protease** inhibitor did not significantly enhance the **resistance** to oxygen. Additionally, neither p53-deficient mice nor lpr mice (Fas null) manifested an increased **resistance** to hyperoxia-induced lung damage. These results show that both necrosis and apoptosis contribute to cell death during hyperoxia. Multiple apoptotic pathways seem to be involved in this, and an antiapoptotic strategy does not attenuate alveolar damage.

L9 ANSWER 20 OF 31 MEDLINE DUPLICATE 11

ACCESSION NUMBER: 1998161865 MEDLINE

DOCUMENT NUMBER: 98161865 PubMed ID: 9495810

TITLE: Caspase activation in MCF7 cells responding to etoposide treatment.

AUTHOR: Benjamin C W; Hiebsch R R; Jones D A

CORPORATE SOURCE: Department of Cardiovascular Pharmacology, Pharmacia and Upjohn Company, Kalamazoo, Michigan 49001, USA..

cwbenjam@am.pnu.com
SOURCE: MOLECULAR PHARMACOLOGY, (1998 Mar) 53 (3) 446-50.
Journal code: NGR; 0035623. ISSN: 0026-895X.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199803
ED Entered STN: 19980410
Last Updated on STN: 20000303
Entered Medline: 19980327

AB Studies of the biochemical mechanisms evoked by conventional treatments for neoplastic diseases point to apoptosis as a key process for elimination of unwanted cells. Although the pathways through which chemotherapeutics promote cell death remain largely unknown, caspase **proteases** play a central role in the induction of apoptosis in response to a variety of stimuli including tumor necrosis factor, **fas ligand**, and growth factor deprivation. In this article, we demonstrate the induction of caspase **protease** activity in MCF7 human breast carcinoma cells exposed to the topoisomerase inhibitor, etoposide. Caspase **protease** activity was assessed by incubating cell lysates with the known caspase substrates, acetyl-L-aspartic-L-glutamic-L-valyl-L-aspartic acid 4-methyl-7-aminocoumarin or acetyl-L-tyrosyl-L-valyl-L-aspartic acid 4-methyl-7-aminocoumarin. We observed maximal cleavage of acetyl-L-aspartic-L-glutamic-L-valyl-L-aspartic acid 4-methyl-7-aminocoumarin within 6 hr following etoposide addition, a time that precedes cell death. In contrast, acetyl-L-tyrosyl-L-valyl-L-aspartic acid 4-methyl-7-aminocoumarin was **resistant** to cleavage activity. This substrate cleavage specificity implies that a caspase-3-like **protease** is activated in response to DNA damage. Consistent with the lysate **protease** activity, an intracellular marker of caspase activation, poly-ADP ribose polymerase (PARP), was cleaved in a concentration- and time-dependent manner after etoposide-treatment. PARP cleavage followed caspase activation and reached maximum cleavage between 12 and 16 hr. Incubation of the cells with the peptidic caspase inhibitor z-valine-alanine-asparagine-CH2F prevented caspase activation, inhibited PARP cleavage, and inhibited cell death. Thus, etoposide killing of MCF7 cells requires a caspase-3-like **protease**.

L9 ANSWER 21 OF 31 MEDLINE DUPLICATE 12
ACCESSION NUMBER: 1998143736 MEDLINE
DOCUMENT NUMBER: 98143736 PubMed ID: 9485194
TITLE: TRAIL/Apo-2-ligand-induced apoptosis in human T cells.
AUTHOR: Jeremias I; Herr I; Boehler T; Debatin K M

CORPORATE SOURCE: Division of Molecular Oncology/Pediatrics, German Cancer Research Center, Heidelberg.
SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (1998 Jan) 28 (1) 143-52.
Journal code: EN5; 1273201. ISSN: 0014-2980.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199803
ED Entered STN: 19980326
Last Updated on STN: 20000303
Entered Medline: 19980319

AB Members of the tumor necrosis factor (TNF) family such as CD95 (APO-1/**Fas**) **ligand** (L) trigger apoptosis in lymphoid cells. Recently, a new member of apoptosis-inducing ligands, TRAIL (TNF-related-apoptosis-inducing-ligand)/Apo-2 ligand, was identified that might act in a similar way. We compared TRAIL and CD95L-induced apoptosis in human lymphoid cells. Expression of TRAIL was found in CD4+ and CD8 T cells following activation, suggesting that TRAIL participates in T cell-mediated induction of apoptosis. Similar to CD95L, TRAIL-induced apoptosis in target cells is mediated by activation of caspases (ICE/Ced-3 **proteases**). However, different human lymphoid cell lines and peripheral T cells differ in sensitivity towards induction of apoptosis by TRAIL and CD95L. In addition, T cells are highly sensitive towards CD95L-induced apoptosis after prolonged activation in vitro, but remain completely **resistant** to TRAIL-induced apoptosis. In contrast, T cells from HIV-1-infected patients previously shown to exhibit increased CD95 sensitivity are even more susceptible towards TRAIL-induced cell death. These data suggest that TRAIL might participate in CD95-independent apoptosis of lymphoid cells and might be involved in deregulated apoptosis in diseases such as leukemias and HIV-1 infection.

L9 ANSWER 22 OF 31 SCISEARCH COPYRIGHT 2001 ISI (R)
ACCESSION NUMBER: 1998:272920 SCISEARCH
THE GENUINE ARTICLE: ZE189
TITLE: Chemosensitivity of solid tumor cells in vitro is related to activation of the CD95 system
AUTHOR: Fulda S; Los M; Friesen C; Debatin K M (Reprint)
CORPORATE SOURCE: UNIV ULM, CHILDRENS HOSP, PRITTWITZSTR 43, D-89075 ULM, GERMANY (Reprint); UNIV HEIDELBERG, CHILDRENS HOSP, HEIDELBERG, GERMANY; GERMAN CANC RES CTR, DIV MOL ONCOL, HEIDELBERG, GERMANY
COUNTRY OF AUTHOR: GERMANY
SOURCE: INTERNATIONAL JOURNAL OF CANCER, (30 MAR 1998) Vol. 76, No. 1, pp. 105-114.

Publisher: WILEY-LISS, DIV JOHN WILEY & SONS INC,
605 THIRD AVE, NEW YORK, NY 10158-0012.
ISSN: 0020-7136.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 28

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB We have identified the CD95 system as a key mediator of chemotherapy-induced apoptosis in leukemia and neuroblastoma cells. Here, we report that sensitivity of various solid tumor cell lines for drug-induced cell death corresponds to activation of the CD95 system, Upon drug treatment, strong induction of CD95 ligand (CD95-L) and caspase activity were found in chemosensitive tumor cells (Hodgkin, Ewing's sarcoma, colon carcinoma and small cell lung carcinoma) but not in tumor cells which responded poorly to drug treatment (breast carcinoma and renal cell carcinoma). Blockade of CD95 using F(ab')(2) anti-CD95 antibody fragments markedly reduced drug-induced apoptosis, suggesting that drug-triggered apoptosis depended on CD95-L/receptor interaction. Moreover, drug treatment induced CD95 expression, thereby increasing sensitivity for CD95-induced apoptosis, Drug-induced apoptosis critically depended on activation of caspases (ICE/Ced-3-like **proteases**) since the broad-spectrum inhibitor of caspases zVAD-fmk strongly reduced drug-mediated apoptosis, The prototype substrate of caspases, poly(ADP-ribose) polymerase, was cleaved upon drug treatment, suggesting that CD95-L triggered autocrine/paracrine death via activation of caspases, Our data suggest that chemosensitivity of solid tumor cells depends on intact apoptosis pathways involving activation of the CD95 system and processing of caspases. Our findings may have important implications for new treatment approaches to increase sensitivity and to overcome **resistance** of solid tumors. (C) 1998 Wiley-Liss, Inc.

L9 ANSWER 23 OF 31 SCISEARCH COPYRIGHT 2001 ISI (R)
ACCESSION NUMBER: 97:820794 SCISEARCH
THE GENUINE ARTICLE: YD358
TITLE: Betulinic acid triggers CD95 (APO-1/Fas)- and p53-independent apoptosis via activation of caspases in neuroectodermal tumors
AUTHOR: Fulda S; Friesen C; Los M; Scaffidi C; Mier W; Benedict M; Nunez G; Krammer P H; Peter M E; Debatin K M (Reprint)
CORPORATE SOURCE: UNIV ULM, CHILDRENS HOSP, PRITTWITZSTR 43, D-89075 ULM, GERMANY (Reprint); UNIV HEIDELBERG, CHILDRENS HOSP, DIV HEMATOL ONCOL, D-69120 HEIDELBERG, GERMANY; GERMAN CANC RES CTR, DIV MOL ONCOL, D-69120 HEIDELBERG, GERMANY; GERMAN CANC RES CTR, DIV

IMMUNOGENET, D-69120 HEIDELBERG, GERMANY; GERMAN CANC RES CTR, DIV MOL TOXICOL, D-69120 HEIDELBERG, GERMANY; UNIV MICHIGAN, SCH MED, DEPT PATHOL, ANN ARBOR, MI 48109
COUNTRY OF AUTHOR: GERMANY; USA
SOURCE: CANCER RESEARCH, (1 NOV 1997) Vol. 57, No. 21, pp. 4956-4964.
Publisher: AMER ASSOC CANCER RESEARCH, PUBLIC LEDGER BLDG, SUITE 816, 150 S. INDEPENDENCE MALL W., PHILADELPHIA, PA 19106.
ISSN: 0008-5472.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; CLIN
LANGUAGE: English
REFERENCE COUNT: 50
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Betulinic acid CBA), a melanoma-specific cytotoxic agent, induced apoptosis in neuroectodermal tumors, such as neuroblastoma, medulloblastoma, and Ewing's sarcoma, representing the most common solid tumors of childhood. BA triggered an apoptosis pathway different from the one previously identified for standard chemotherapeutic drugs. BA-induced apoptosis was independent of CD95-ligand/receptor interaction and accumulation of wild-type p53 protein, but it critically depended on activation of caspases (interleukin 1 beta-converting enzyme/Ced-3-like **proteases**), FLICE/MACH (caspase-8), considered to be an upstream **protease** in the caspase cascade, and the downstream caspase CPP32/YAMA/Apopain (caspase-3) were activated, resulting in cleavage of the prototype substrate of caspases PARP. The broad-spectrum peptide inhibitor benzyloxycarbonyl-Val-Ala-Asp-fluoromethylketone, which blocked cleavage of FLICE and PARP, also completely abrogated BA-triggered apoptosis. Cleavage of caspases was preceded by disturbance of mitochondrial membrane potential and by generation of reactive oxygen species. Overexpression of Bcl-2 and Bcl-x(L) conferred **resistance** to BA at the level of mitochondrial dysfunction, **protease** activation, and nuclear fragmentation. This suggested that mitochondrial alterations were involved in BA-induced activation of caspases. Furthermore, pax and Bcl-x(s), two death-promoting proteins of the Bcl-2 family, were up-regulated following BA treatment. Most importantly, neuroblastoma cells **resistant** to CD95- and doxorubicin-mediated apoptosis were sensitive to treatment with BA, suggesting that BA may bypass some forms of drug **resistance**. Because BA exhibited significant antitumor activity on patients' derived neuroblastoma cells ex vivo, BA may be a promising new agent for the treatment of neuroectodermal tumors in vivo.

L9 ANSWER 24 OF 31 MEDLINE DUPLICATE 13

ACCESSION NUMBER: 97433125 MEDLINE
DOCUMENT NUMBER: 97433125 PubMed ID: 9288794
TITLE: The CD95 (APO-1/Fas) system mediates drug-induced apoptosis in neuroblastoma cells.
AUTHOR: Fulda S; Sieverts H; Friesen C; Herr I; Debatin K M
CORPORATE SOURCE: Division of Hematology/Oncology, University Children's Hospital, German Cancer Research Center, Heidelberg.
SOURCE: CANCER RESEARCH, (1997 Sep 1) 57 (17) 3823-9.
Journal code: CNF; 2984705R. ISSN: 0008-5472.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199709
ED Entered STN: 19971008
Last Updated on STN: 20000303
Entered Medline: 19970924

AB Anticancer agents have been shown to trigger apoptosis in chemosensitive tumors such as neuroblastomas. We previously identified activation of the CD95 system as one of the key mechanisms for doxorubicin-induced apoptosis in leukemic T cells. Here, we report that therapeutic concentrations of doxorubicin, cisplatinum, and VP-16 led to induction of CD95 receptor and CD95 ligand (CD95-L) that mediated cell death in chemosensitive neuroblastoma cells. Using F(ab')2 anti-CD95 antibody fragments to interfere with CD95-L-receptor interaction markedly reduced apoptosis induced by those drugs in vitro. Cyclosporin A inhibited induction of CD95 mRNA and CD95-L mRNA and blocked drug-mediated apoptosis. Drug-induced apoptosis involved activation of caspases (interleukin 1beta-converting enzyme/Ced-3-like **proteases**) and processing of the prototype caspase substrate PARP and was completely blocked by benzyloxycarbonyl-Val-Ala-Asp-fluoromethyl ketone, a peptide inhibitor of caspases. In addition, neuroblastoma cells that were **resistant** to CD95-triggered apoptosis also displayed cross-**resistance** to chemotherapeutic agents. These data provide new clues for understanding the molecular requirements for drug-induced apoptosis in chemosensitive neuroblastoma cells by demonstrating that cell death was mediated via the CD95-L-receptor system and may open new avenues for targeting drug **resistance** of neuroblastoma.

L9 ANSWER 25 OF 31 MEDLINE DUPLICATE 14
ACCESSION NUMBER: 97461432 MEDLINE
DOCUMENT NUMBER: 97461432 PubMed ID: 9317113
TITLE: Induction of T cell clonal anergy results in resistance, whereas CD28-mediated costimulation primes for susceptibility to Fas- and Bax-mediated

programmed cell death.

AUTHOR: Boussiotis V A; Lee B J; Freeman G J; Gribben J G; Nadler L M
CORPORATE SOURCE: Department of Medicine, Harvard Medical School, Boston, MA 02115, USA.

CONTRACT NUMBER: AI-35225-04 (NIAID)
CA-40216-12 (NCI)
SOURCE: JOURNAL OF IMMUNOLOGY, (1997 Oct 1) 159 (7) 3156-67.
Journal code: IFB; 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199710
ED Entered STN: 19971105
Last Updated on STN: 20000303
Entered Medline: 19971021

AB Since TCR-mediated stimulation induces T cells to become sensitive to Fas-mediated activation-induced cell death (Fas-AICD), we examined whether anergized and CD28-costimulated T cell clones were equally sensitive to Fas-AICD. Here, we show that TCR signal in the presence or absence of CD28 costimulation induced equivalent expression of Fas and **Fas ligand**. Although anergized cells expressed Fas and **Fas ligand**, they were **resistant** to Fas-AICD. Induction of anergy resulted in up-regulation and persistent expression of moderate amounts of bcl-xL and bax and absence of induction of bad. In contrast, CD28-costimulated cells that also expressed Fas and **Fas ligand** were initially **resistant** to Fas-AICD but became susceptible after 72 h of culture. Although Fas-mediated apoptosis was the major mechanism of AICD, the IL-1beta-converting enzyme-like **protease** inhibitor zVAD-FMK totally abrogated DNA fragmentation but not cell death, suggesting that additional Fas-independent apoptotic mechanisms were also operative. **Resistance** to apoptotic cell death was temporally associated with a dramatic increase of bcl-xL and the presence of bcl-xL:bax heterodimers. Subsequent sensitivity to AICD was associated with down-regulation of bcl-xL, induction of bad, and the displacement of bax from bcl-xL:bax heterodimers. Although induced following CD28 costimulation, bcl-2 did not protect against AICD. Therefore, besides its role in promotion of viability, prevention of anergy, and clonal expansion, CD28 costimulation also has a central role in the induction of subsequent AICD by up-regulating apoptotic mediators.

L9 ANSWER 26 OF 31 MEDLINE
ACCESSION NUMBER: 1998025896 MEDLINE
DOCUMENT NUMBER: 98025896 PubMed ID: 9376593

TITLE: Cross-**resistance** of CD95- and drug-induced apoptosis as a consequence of deficient activation of caspases (ICE/Ced-3 **proteases**).
AUTHOR: Los M; Herr I; Friesen C; Fulda S; Schulze-Osthoff K; Debatin K M
CORPORATE SOURCE: Hematology/Oncology, University Children's Hospital, Ulm, Germany.
SOURCE: BLOOD, (1997 Oct 15) 90 (8) 3118-29.
Journal code: A8G; 7603509. ISSN: 0006-4971.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199711
ED Entered STN: 19971224
Last Updated on STN: 20000303
Entered Medline: 19971112

AB The cytotoxic effect of anticancer drugs has been shown to involve induction of apoptosis. We report here that tumor cells **resistant** to CD95 (APO-1/Fas) -mediated apoptosis were cross-**resistant** to apoptosis-induced by anticancer drugs. Apoptosis induced in tumor cells by cytarabine, doxorubicin, and methotrexate required the activation of ICE/Ced-3 **proteases** (caspases), similarly to the CD95 system. After drug treatment, a strong increase of caspase activity was found that preceded cell death. Drug-induced activation of caspases was also found in ex vivo-derived T-cell leukemia cells. **Resistance** to cell death was conferred by a peptide caspase inhibitor and CrmA, a poxvirus-derived serpin. The peptide inhibitor was effective even if added several hours after drug treatment, indicating a direct involvement of caspases in the execution and not in the trigger phase of drug action. Drug-induced apoptosis was also strongly inhibited by antisense approaches targeting caspase-1 and -3, indicating that several members of this **protease** family were involved. CD95-**resistant** cell lines that failed to activate caspases upon CD95 triggering were cross-**resistant** to drug-mediated apoptosis. Our data strongly support the concept that sensitivity for drug-induced cell death depends on intact apoptosis pathways leading to activation of caspases. The identification of defects in caspase activation may provide molecular targets to overcome drug **resistance** in tumor cells.

L9 ANSWER 27 OF 31 MEDLINE DUPLICATE 15
ACCESSION NUMBER: 97349047 MEDLINE
DOCUMENT NUMBER: 97349047 PubMed ID: 9205051
TITLE: p53-dependent DNA damage-induced apoptosis requires Fas/APO-1-independent activation of CPP32beta.

AUTHOR: Fuchs E J; McKenna K A; Bedi A
CORPORATE SOURCE: Johns Hopkins Oncology Center, Johns Hopkins University School of Medicine, Baltimore, Maryland 21287, USA.
CONTRACT NUMBER: K08AI01249 (NIAID)
R29CA71660-01A1 (NCI)
SOURCE: CANCER RESEARCH, (1997 Jul 1) 57 (13) 2550-4.
Journal code: CNF; 2984705R. ISSN: 0008-5472.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199707
ED Entered STN: 19970812
Last Updated on STN: 20000303
Entered Medline: 19970725

AB In many cell types, the p53 tumor suppressor protein is required for the induction of apoptosis by DNA-damaging chemotherapy or radiation. Therefore, identification of the molecular determinants of p53-dependent cell death may aid in the design of effective therapies of p53-deficient cancers. We investigated whether p53-dependent apoptosis requires activation of CPP32beta (caspase 3), a cysteine **protease** that has been found to mediate apoptosis in response to ligation of the Fas molecule or to granzyme B, a component of CTL lytic granules. Irradiation-induced apoptosis was associated with p53-dependent activation of CPP32beta-related proteolysis, and normal thymocytes were protected from irradiation by Acetyl-Asp-Glu-Val-Asp-CHO (Ac-DEVD-CHO), a specific inhibitor of CPP32beta. We next examined whether the Fas system is required for p53-dependent apoptosis and whether stimuli that induce activation of CPP32beta induce apoptosis in p53-deficient cells. Thymocytes or activated T cells from Fas-deficient mice were **resistant** to apoptosis induced by ligation of Fas or CD3, respectively, but remained normally susceptible to irradiation. Thymocytes from p53-deficient mice, although **resistant** to DNA damage, remained sensitive to CPP32beta-mediated apoptosis induced by ligation of Fas or CD3, or by exposure to cytotoxic T cells. These results demonstrate that DNA damage-induced apoptosis of T cells requires p53-mediated activation of CPP32beta by a mechanism independent of Fas/**FasL** interactions and suggest that immunological or molecular methods of activating CPP32beta may be effective at inducing apoptosis in p53-deficient cancers that are **resistant** to conventional chemotherapy or irradiation.

L9 ANSWER 28 OF 31 SCISEARCH COPYRIGHT 2001 ISI (R)
ACCESSION NUMBER: 97:389296 SCISEARCH
THE GENUINE ARTICLE: WY883
TITLE: ICE-proteases mediate HTLV-I tax-induced apoptotic

T-cell death

AUTHOR: Chlichlia K; Busslinger M; Peter M E; Walczak H; Krammer P H; Schirrmacher V; Khazaie K (Reprint)

CORPORATE SOURCE: GERMAN CANC RES CTR, TUMORIMMUNOL PROGRAM, NEUENHEIMER FELD 280, D-69120 HEIDELBERG, GERMANY (Reprint); GERMAN CANC RES CTR, TUMORIMMUNOL PROGRAM, D-69120 HEIDELBERG, GERMANY; RES INST MOL PATHOL, A-1030 VIENNA, AUSTRIA; UNIV HEIDELBERG, DEPT SURG, SECT MOL DIAGNOST & THERAPY, D-69120 HEIDELBERG, GERMANY

COUNTRY OF AUTHOR: GERMANY; AUSTRIA

SOURCE: ONCOGENE, (15 MAY 1997) Vol. 14, No. 19, pp. 2265-2272.
Publisher: STOCKTON PRESS, HOUNDMILLS, BASINGSTOKE, HAMPSHIRE, ENGLAND RG21 6XS.
ISSN: 0950-9232.

DOCUMENT TYPE: Article; Journal

FILE SEGMENT: LIFE

LANGUAGE: English

REFERENCE COUNT: 50

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The Tax protein of Human T-cell leukemia virus type 1 (HTLV-1) is important for the T-cell immortalizing properties of this virus in vitro and is considered to be responsible for the early stages of leukemogenesis in infected hosts, Tax can upregulate expression of TNF-alpha and TNF-beta, as well as potentiate apoptosis in activated T-cells and in serum starved murine fibroblasts. To examine the role of CD95 (APO-1/Fas) and ICE-**proteases** in Tax-mediated active T-cell death, Jurkat T cells expressing (APO(S)) or lacking (APO(R)) cell surface expression of CD95 (APO-1/Fas) were genetically modified to express hormone-inducible HTLV-1 Tax constructs, Hormone-inducible action of Tax alone was sufficient to promote programmed cell death in CD95-expressing Jurkat T-cell clones, In contrast, clones lacking CD95 surface expression were **resistant** to the antiproliferative action of Tax, Both APOS and APOR clones exhibited Tax-dependent upregulation of CD95 ligand and TNF-alpha. Blocking experiments suggested that while the apoptotic action of Tax critically required ICE-**protease** function it was largely independent of cell surface interaction of CD95 Ligand or TNF-alpha with their corresponding receptors, These observations strongly implicate ICE-**proteases** in Tax-induced T-cell death, and suggest a possible involvement of CD95 in this process.

L9 ANSWER 29 OF 31 MEDLINE DUPLICATE 16

ACCESSION NUMBER: 97385026 MEDLINE
DOCUMENT NUMBER: 97385026 PubMed ID: 9242521

TITLE: Comparison of apoptosis in wild-type and

Fas-resistant cells: chemotherapy-induced apoptosis is not dependent on Fas/**Fas ligand** interactions.

AUTHOR: Eischen C M; Kottke T J; Martins L M; Basi G S; Tung J S; Earnshaw W C; Leibson P J; Kaufmann S H

CORPORATE SOURCE: Department of Immunology, Mayo Clinic, Rochester, MN 55901, USA.

CONTRACT NUMBER: CA69008 (NCI)

SOURCE: BLOOD, (1997 Aug 1) 90 (3) 935-43.
Journal code: A8G; 7603509. ISSN: 0006-4971.

PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals

ENTRY MONTH: 199708

ED Entered STN: 19970908
Last Updated on STN: 20000303
Entered Medline: 19970825

AB The Fas/**Fas ligand** (**FasL**) pathway is widely involved in apoptotic cell death in lymphoid and nonlymphoid cells. It has recently been postulated that many chemotherapeutic agents also induce cell death by activating the Fas/**FasL** pathway. In the present study we compared apoptotic pathways induced by anti-Fas or chemotherapeutic agents in the Jurkat human T-cell leukemia line. Immunoblotting showed that treatment of wild-type Jurkat cells with anti-Fas or the topoisomerase II-directed agent etoposide resulted in proteolytic cleavage of precursors for the cysteine-dependent aspartate-directed **proteases** caspase-3 and caspase-7 and degradation of the caspase substrates poly(ADP-ribose) polymerase (PARP) and lamin B1. Likewise, affinity labeling with N-(N(alpha)-benzyloxycarbonylglutamyl-N(epsilon)-biotinyllysyl+ ++)aspartic acid [(2,6-dimethyl-benzoyl)oxy]methyl ketone [Z-EK(bio)D-amok] labeled the same five active caspase species after each treatment, suggesting that the same downstream apoptotic pathways have been activated by anti-Fas and etoposide. Treatment with ZB4, an antibody that inhibits Fas-mediated cell death, failed to block etoposide-induced apoptosis, raising the possibility that etoposide does not initiate apoptosis through Fas/**FasL** interactions. To further explore the relationship between Fas- and chemotherapy-induced apoptosis, Fas-**resistant** Jurkat cells were treated with various chemotherapeutic agents. Multiple independently derived Fas-**resistant** Jurkat lines underwent apoptosis that was indistinguishable from that of the Fas-sensitive parental cells after treatment with etoposide, doxorubicin, topotecan, cisplatin, methotrexate, staurosporine, or gamma-irradiation. These results indicate that antineoplastic treatments induce apoptosis through a Fas-independent pathway even though Fas- and chemotherapy-induced

pathways converge on common downstream apoptotic effector molecules.

L9 ANSWER 30 OF 31 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1997:328296 BIOSIS
DOCUMENT NUMBER: PREV199799627499
TITLE: **Resistance** to apoptosis induced by TNF-alpha, but not **fas ligand**, in human leukaemic cells is conferred by a protein which acts upstream of ICE/ICE-like **protease** activation.
AUTHOR(S): Patel, S.; Allen, P. D.; Kelsey, S. M.; Newland, A. C.
CORPORATE SOURCE: Dep. Haematol., St. Bartholomews and Royal London Hosp. Sch. Med. Dentistry, Turner St., London E1 2AD UK
SOURCE: British Journal of Haematology, (1997) Vol. 97, No. SUPPL. 1, pp. 88.
Meeting Info.: Annual Scientific Meeting of the British Society for Haematology Harrogate, England, UK April 14-17, 1997
ISSN: 0007-1048.
DOCUMENT TYPE: Conference; Abstract
LANGUAGE: English

L9 ANSWER 31 OF 31 SCISEARCH COPYRIGHT 2001 ISI (R)
ACCESSION NUMBER: 1998:221116 SCISEARCH
THE GENUINE ARTICLE: ZB486
TITLE: Apoptosis and its role in tumor growth.
AUTHOR: Vladimirskaya E B (Reprint); Maschan A A; Rumyantsev A G
CORPORATE SOURCE: PEDIAT HEMATOL RES INST, MOSCOW, RUSSIA (Reprint)
COUNTRY OF AUTHOR: RUSSIA
SOURCE: GEMATOLOGIYA I TRANSFUZIOLOGIYA, (SEP-OCT 1997) Vol. 42, No. 5, pp. 4-9.
Publisher: MINISTERSTVO ZDRAVOOKHRANENIYA, NAUCHNIY PROEZD 6, B-246 MOSCOW, RUSSIA.
ISSN: 0234-5730.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: CLIN
LANGUAGE: Russian
REFERENCE COUNT: 89
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Molecular genetic investigations show that in addition to BCL-2 genes mapped in humans on chromosome 18 there are other genes responsible for enhancement or inhibition or apoptosis (BAX, BAK, BAD, P-53, C-MYC, APO-1/FAS and C-FES, respectively). Growth factors, extracellular matrix, CD-40-ligand, neutral amino acids, zink, estrogens, androgens belong to basic physiological inhibitors

of apoptosis. Growth factors are the most potent antiapoptotic stimuli for normal hemopoiesis. Cytokines SF, TPO, EPO, GM-CSF, G-CSF, M-CSF, IL-2, IL-3, IL-4, IL-10, alpha-IFN prevent apoptosis while such cytokines as IL-I, IL-4, IL-10, gamma-IFN, TFG and TNF induce apoptosis. Antitumor drugs are apoptosis inducers, i.e. they cause death of tumor cells. The pattern of apoptosis in this case is similar to that used by physiological apoptosis inducers: stimulation of P-53 gene, modulation of BCL-2 genes, inclusion of FAS-receptor, activation of effector **proteases** and endonucleases, blocking of receptors of antiapoptotic growth factors and stimulation of proapoptotic growth factors. Common reason of **resistance** of tumor cells to drugs is mutation or deficiency of gene P-53 or overexpression of gene BCL-2 making cells unresponsible to all the proapoptotic stimuli. The study of apoptosis mechanisms and methods of their regulation enables new interpretation of hemopoiesis, oncogenesis events, open perspectives for advancements in tumor treatment.

FasL as treatment

```
     (FILE 'CAPLUS' ENTERED AT 10:40:14 ON 26 APR 2001)
L4             8 SEA FILE=REGISTRY ABB=ON  PLU=ON  FAS LIGAND ?/CN
L5          2989 SEA FILE=CAPLUS ABB=ON  PLU=ON  FAS(W)(L OR LIGAND) OR
                 FASL OR L4
L14           27 SEA FILE=CAPLUS ABB=ON  PLU=ON  L5(5A)ADMIN?
L18       140108 SEA FILE=CAPLUS ABB=ON  PLU=ON  (TREAT? OR THERAP?)(10A)(
                 DISEAS? OR DISORDER OR CONDITION)
L19            2 SEA FILE=CAPLUS ABB=ON  PLU=ON  L14 AND L18

L20            2 L19 NOT L7

=> d 1-2 .bevstr

L20  ANSWER 1 OF 2  CAPLUS  COPYRIGHT 2001 ACS
ACCESSION NUMBER:         1998:433887  CAPLUS
DOCUMENT NUMBER:          129:147989
TITLE:                    Involvement of Fas-mediated apoptosis in the
                          hematopoietic progenitor cells of
                          graft-versus-host reaction-associated
                          myelosuppression
AUTHOR(S):                Mori, Takehiko; Nishimura, Takashi; Ikeda,
                          Yasuo; Hotta, Tomomitsu; Yagita, Hideo; Ando,
                          Kiyoshi
CORPORATE SOURCE:         Division of Hematology, The Departments of
                          Internal Medicine and Immunology, Research
                          Center for Genetic Engineering and Cell
                          Transplantation, Tokai University School of
                          Medicine, Kanagawa, 259-1193, Japan
SOURCE:                   Blood (1998), 92(1), 101-107
                          CODEN: BLOOAW; ISSN: 0006-4971
```

PUBLISHER: W. B. Saunders Co.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The influence of graft-vs.-host (GVH) reaction on the host hematopoietic cells clin. manifests itself both as adverse reactions in transfusion-assocd. GVH **disease** (GVHD) and as a **therapeutic** graft-vs.-leukemia (GVL) effect in either donor lymphocytes transfusion (DLT) or allogeneic bone marrow (BM) transplantation. We examd. the effect of GVH reaction on the host hematopoiesis in the murine parent-into-F1 (P1 .fwdarw. F1) model of GVHD. The systemic transfer of 5.times.107 of C57BL/6 (B6) splenocytes into (B6xDBA/2)F1 mice (BDF1), which results in acute GVHD, reduced the peripheral blood cell counts, the no. of BM cells, and colony-forming unit-granulocyte macrophage (CFU-GM), whereas the injection of 108 of DBA/2 cells into BDF1, which results in chronic GVHD, did not affect hematopoiesis 2 wk after the transfer. To clarify the mechanism of such myelosuppression, we examd. the Fas expression in both hematopoietic progenitor cells as well as whole BM cells. The Fas expressions in each fraction significantly increased in BDF1 mice 2 wk after the induction of acute GVHD, whereas no such effects were obsd. in the BDF1 mice with chronic GVHD. Furthermore, when such BM cells were incubated with anti-Fas antibody (Jo2), which induces apoptosis through Fas, the fraction of apoptotic cells increased and the no. of CFU-GM decreased significantly. The in vivo **administration** of neutralizing anti-**FasL** antibody into BDF1 mice receiving with B6 spleen cells thus protected the host mice from BM failure. These results indicate that the functional expression of Fas on hematopoietic cells plays an essential role in the myelosuppressive effect of GVHD.

L20 ANSWER 2 OF 2 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:310806 CAPLUS
DOCUMENT NUMBER: 126:288116
TITLE: Methods for **treatment** of **diseases** associated with a deficiency of Fas ligand activity
INVENTOR(S): Kaplan, David R.
PATENT ASSIGNEE(S): Tkb Associates Limited Partnership, USA
SOURCE: PCT Int. Appl., 54 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE

WO 9712632 A1 19970410 WO 1996-US15917 19961004
W: CA, JP
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE
PRIORITY APPLN. INFO.: US 1995-4985 19951005
AB Methods for **treating** a patient with a **condition** characterized by a deficiency of **Fas ligand** activity comprise **administering** an agent that increases the **Fas ligand** activity in the patient. The agent is e.g. an anti-Fas antibody, e.g. a bispecific antibody having a Fas antigen domain and a cell-surface marker-specific domain. Also disclosed is a method for treating a viral infection by administering an agent that increases apoptosis of Fas+ cells. Results are presented which demonstrate that peripheral blood mononuclear cells (PBMC) from HIV-infected patients were depressed in their capacity to mediate Fas ligand activity, as compared to PBMC from healthy volunteers. Anti-Fas IgM was shown to inhibit HIV prodn. Inhibition of Fas ligand activity by herpes simplex virus type 2 is also described.

(FILE 'MEDLINE, BIOSIS, EMBASE, WPIDS, CONFSCI, SCISEARCH, JICST-EPLUS, JAPIO' ENTERED AT 10:45:29 ON 26 APR 2001)
L21 10 S L19
L22 10 S L21 NOT L8
L23 7 DUP REM L22 (3 DUPLICATES REMOVED)

L23 ANSWER 1 OF 7 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1999-494217 [41] WPIDS
DOC. NO. CPI: C1999-144833
TITLE: Preventives and remedies containing anti-Fas ligand antibody as active ingredient, for inflammatory intestinal disease, ischemic colitis and/or idiopathic inflammatory intestinal diseases .
DERWENT CLASS: B04 D16
INVENTOR(S): NAGATA, S; SUDA, T; YATOMI, T
PATENT ASSIGNEE(S): (MOCH) MOCHIDA PHARM CO LTD; (OSAB-N) OSAKA BIOSCIENCE INST
COUNTRY COUNT: 22
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9939737	A1	19990812	(199941)*	JA	32
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE					
W: CA JP US					
EP 1059089	A1	20001213	(200066)	EN	
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE					

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9939737	A1	WO 1999-JP496	19990205
EP 1059089	A1	EP 1999-902839	19990205
		WO 1999-JP496	19990205

FILING DETAILS:

PATENT NO	KIND		PATENT NO
EP 1059089	A1	Based on	WO 9939737

PRIORITY APPLN. INFO: JP 1998-25492 19980206
AN 1999-494217 [41] WPIDS
AB WO 9939737 A UPAB: 19991011
NOVELTY - An anti-Fas ligand antibody remedy for preventing or **treating** inflammatory intestinal **disease**, ischemic colitis and idiopathic inflammatory intestinal **disease** induced by infection, chemicals and radiation, is new.
ACTIVITY - Apoptosis-inhibitor; anti-inflammatory; anti-inflammation.
MECHANISM OF ACTION - Apoptosis inhibition; Fas-Fas ligand biological effect.
USE - The agent can be used to **treat** or prevent 1 or more of inflammatory intestinal **disease**, ischemic colitis or Crohn's disease and ulcerative colitis and idiopathic inflammatory intestinal disease induced by infection, chemicals and radiation (claimed), with apoptosis inhibition, particularly due to Fas-involved apoptosis. The **therapeutic** efficacy during the active period of the **disease** improves recovery from mucosal lesions, healing of the gastrointestinal tract, or/and relieving diarrhea.
DESCRIPTION OF DRAWING(S) - The diagram shows the effect of anti-mouse Fas ligand antibody on improving disease state in inflammatory colitis model:
white column = control group;
black columns = anti-mouse **Fas ligand** antibody **administration** groups.
Dwg.1/2

L23 ANSWER 2 OF 7 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1999-132243 [11] WPIDS
CROSS REFERENCE: 1999-132242 [11]
DOC. NO. NON-CPI: N1999-096267
DOC. NO. CPI: C1999-038774
TITLE: Inhibition of proinflammatory responses - using an

agent which modulates FasL stimulation, used for **treating** graft versus host **disease** or autoimmune **disease**.

DERWENT CLASS: B04 D16 S03
INVENTOR(S): CHEN, J; NABEL, G J
PATENT ASSIGNEE(S): (UNMI) UNIV MICHIGAN
COUNTRY COUNT: 82
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9903999	A1	19990128	(199911)*	EN	70

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SZ UG ZW
W: AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GE GH GM HR HU ID IL IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG US UZ VN YU ZW

AU 9884087	A	19990210	(199925)		

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9903999	A1	WO 1998-US14771	19980716
AU 9884087	A	AU 1998-84087	19980716

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 9884087	A	Based on	WO 9903999

PRIORITY APPLN. INFO: US 1997-52829 19970717
AN 1999-132243 [11] WPIDS
CR 1999-132242 [11]
AB WO 9903999 A UPAB: 19990316

A method is claimed for inhibiting a proinflammatory response in a suitable cell mixture comprising administering to the mixture an agent which suppresses a FasL proinflammatory response.

Also claimed are:

(1) a method of inhibiting a FasL-mediated proinflammatory response in a subject comprising administering an immunosuppressive agent that specifically inhibits the proinflammatory effect of FasL;

(2) a method for identifying agents which modulate FasL stimulation of a proinflammatory response, comprising: (a) contacting a target cell mixture and a control cell mixture with FasL and an immunosuppressive agent; (b) contacting the target cell

mixture with a candidate therapeutic agent; (c) assaying the target cell mixture for localised proinflammatory response; and (d) comparing the target cell mixture to the control cell mixture to determine if the agent modulates localised FasL stimulation of the proinflammatory response;

(3) a method for identifying agents which modulate FasL stimulation of a proinflammatory response, comprising: (a) contacting a target cell mixture and a control cell mixture with FasL; (b) contacting the target cell mixture with a candidate therapeutic agent and the control cell mixture with an immunosuppresive agent; (c) assaying the target cell mixture for localised proinflammatory response; and (d) comparing the target cell mixture to the control cell mixture to determine if the agent modulates localised FasL stimulation of the proinflammatory response;

(4) a method for identifying agents which modulate FasL stimulation of a proinflammatory response, comprising: (a) contacting a target cell mixture and a control cell mixture with an immunosuppressive agent; (b) contacting the target cell mixture with a candidate therapeutic agent and the control cell mixture with FasL; (c) assaying the target cell mixture for localised proinflammatory response; and (d) comparing the target cell mixture to the control cell mixture to determine if the agent modulates FasL-mediated stimulation of the proinflammatory response;

(5) a method for inhibiting the **FasL**-mediated proinflammatory response by **administering** an agent which stimulates the production of transforming growth factor (TGF)- beta 1-5.

USE - The method can be used for **treating diseases** associated with an undesired FasL-mediated proinflammatory response, e.g. graft versus host disease, or an autoimmune disease such as systemic lupus erythematosus, rheumatoid arthritis and psoriasis.

Dwg.0/8

L23 ANSWER 3 OF 7 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1999:376363 BIOSIS
DOCUMENT NUMBER: PREV199900376363
TITLE: Treatment of experimental glioma by **administration** of adenoviral vectors expressing **Fas ligand**.
AUTHOR(S): Ambar, Benjamin B.; Frei, Karl; Malipiero, Ursula; Morelli, Adrian E.; Castro, Maria G.; Lowenstein, Pedro R.; Fontana, Adriano (1)
CORPORATE SOURCE: (1) Section of Clinical Immunology, University Hospital Zurich, Haldeliweg 4, 8044, Zurich Switzerland
SOURCE: Human Gene Therapy, (July 1, 1999) Vol. 10, No. 10,

pp. 1641-1648.
ISSN: 1043-0342.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Fas ligand (FasL) is a cytokine, produced by activated T cells and NK cells, that triggers apoptosis of Fas-positive target cells including human glioma cells. As shown here, in vitro infection of rat F98 and human LN18 glioma cell lines with recombinant adenovirus (rAd) expressing FasL cDNA under control of the cytomegalovirus promoter (rAd-CMV-FasL) induced striking cytotoxicity in Fas-positive glioma cell lines but not in the Fas-negative F98 glioma subline F98/ZH. The extent of FasL-mediated cytotoxic effects outranged the expectations based on expression of beta-galactosidase (beta-Gal) by F98 cells infected with a control virus expressing the lacZ gene (rAd-CMV-lacZ). The detection of FasL bioactivity in supernatants of infected cells provides evidence of a bystander mechanism involving the cytotoxic action of FasL on uninfected cells. In F98 tumor-bearing rats, infection with rAd-CMV-FasL increased the mean survival time by 50% compared with infection with rAd-CMV-lacZ or untreatedcontrols. These data suggest that viral vector transduction of the FasL gene could be part of a successful glioma gene therapy.

L23 ANSWER 4 OF 7 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1999:343369 BIOSIS
DOCUMENT NUMBER: PREV199900343369
TITLE: Gene therapy of experimental autoimmune thyroiditis by in vivo **administration** of plasmid DNA coding for **Fas ligand** or IL-10.
AUTHOR(S): Chiocchia, Gilles; Batteux, Frederic; Tourneur, Lea; Trebeden, Helene; Charreire, Jeannine
SOURCE: Journal of Autoimmunity, (1999) No. SUPPL., pp. 2. Meeting Info.: 2nd International Congress on Autoimmunity Tel Aviv, Israel March 7-11, 1999
ISSN: 0896-8411.
DOCUMENT TYPE: Conference
LANGUAGE: English

L23 ANSWER 5 OF 7 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1998-506345 [43] WPIDS
DOC. NO. CPI: C1998-152776
TITLE: Use of Fas ligand molecules - for **treating conditions** characterised by excessive proliferation of smooth muscle cells, e.g. arteriosclerosis, vascular injury or reperfusion.
DERWENT CLASS: B04 D16
INVENTOR(S): WALSH, K

PATENT ASSIGNEE(S): (SELI-N) ST ELIZABETH'S MEDICAL CENT BOSTON INC;
(SELI-N) ST ELIZABETH'S MEDICAL CENT
COUNTRY COUNT: 22
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9839354	A1	19980911	(199843)*	EN	75
RW: AT BE CH DE DK ES FI FR GB GR IE IT LU MC NL PT SE					
W: AU CA JP					
AU 9863403	A	19980922	(199908)		
US 5858990	A	19990112	(199910)		
EP 981536	A1	20000301	(200016)	EN	
R: AT BE CH DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE					

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9839354	A1	WO 1998-US3781	19980226
AU 9863403	A	AU 1998-63403	19980226
US 5858990	A	US 1997-810453	19970304
EP 981536	A1	EP 1998-907648	19980226
		WO 1998-US3781	19980226

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 9863403	A	Based on	WO 9839354
EP 981536	A1	Based on	WO 9839354

PRIORITY APPLN. INFO: US 1997-810453 19970304
AN 1998-506345 [43] WPIDS
AB WO 9839354 A UPAB: 19981203
The following are claimed: (A) a method for **treating** a **condition** characterised by excessive proliferation of smooth muscle cells (SMCs) in a subject otherwise free of symptoms calling for **Fas ligand** molecule treatment, by **administering** a **Fas ligand** molecule to a subject to inhibit excessive SMC proliferation; (B) a method for treating a subject who has sustained a vascular injury, comprising **administering** a **Fas ligand** molecule to a subject to inhibit vascular SMC proliferation, where the subject is otherwise free of symptoms calling for Fas ligand molecule treatment; (C) a method for inhibiting vascular remodelling in a subject otherwise free of symptoms calling for **Fas ligand** treatment, comprising **administering** a

Fas ligand molecule to a subject to inhibit vascular remodelling, where the subject is otherwise free of symptoms call for treatment with a Fas ligand molecule; (D) an isolated nucleic acid molecule (NAM): (a) which hybridises under stringent conditions to a molecule consisting of the nucleic acids sequence shown and which codes for a membrane-associated Fas ligand polypeptide; (b) nucleic acid molecules that differ from the NAMs as in (a) in codon sequence due to the degeneracy of the genetic code, and (c) complements of (a) and (b); (E) a vector comprising an isolated NAM as in (D), operably linked to a promoter; (F) a host cell transformed or transfected with an expression vector comprising an isolated NAM as in (D), operably linked to a promoter; (G) an isolated polypeptide coded for by an isolated NAM as in (D); (H) an isolated NAM: (a) which hybridises under stringent conditions to a molecule consisting of the nucleic acid sequence comprising 1790 bp (given in the specification), and which codes for an intact Fas ligand polypeptide or a membrane-associated Fas ligand polypeptide which cannot be cleaved in vivo to provide a soluble Fas ligand polypeptide; (b) NAMs that differ from the NAMs as in (a) in codon sequence due to the degeneracy of the genetic code; and (c) complements of (a) and (b); (I) a vector comprising an isolated NAM as in (H), operably linked to a promoter; (J) a host cell transformed or transfected with an expression vector comprising an isolated NAM as in (H), operably linked to a promoter, and (K) an isolated polypeptide coded for by an isolated NAM as in (H).

USE - The Fas ligand molecules can induce apoptosis in SMCs. They can be used for treating or preventing e.g. arteriosclerosis, including atherosclerosis and post interventional restenosis or other vessel wall injury-induced excessive vascular remodelling and cardio-vascular remodelling, vein graft occlusion, ischemic heart disease or pulmonary hypertension.

Dwg.0/0

L23 ANSWER 6 OF 7 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 1998303750 MEDLINE
DOCUMENT NUMBER: 98303750 PubMed ID: 9639505
TITLE: Involvement of Fas-mediated apoptosis in the hematopoietic progenitor cells of graft-versus-host reaction-associated myelosuppression.
AUTHOR: Mori T; Nishimura T; Ikeda Y; Hotta T; Yagita H; Ando K
CORPORATE SOURCE: Division of Hematology, the Departments of Internal Medicine and Immunology, Research Center for Genetic Engineering and Cell Transplantation, Tokai University School of Medicine, Kanagawa, Japan.
SOURCE: BLOOD, (1998 Jul 1) 92 (1) 101-7.
Journal code: A8G; 7603509. ISSN: 0006-4971.
PUB. COUNTRY: United States

Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199807
ED Entered STN: 19980731
Last Updated on STN: 19980731
Entered Medline: 19980721

AB The influence of graft-versus-host (GVH) reaction on the host hematopoietic cells clinically manifests itself both as adverse reactions in transfusion-associated GVH **disease** (GVHD) and as a **therapeutic** graft-versus-leukemia (GVL) effect in either donor lymphocytes transfusion (DLT) or allogeneic bone marrow (BM) transplantation. We examined the effect of GVH reaction on the host hematopoiesis in the murine parent-into-F1 (P1 --> F1) model of GVHD. The systemic transfer of 5 x 10(7) of C57BL/6 (B6) splenocytes into (B6xDBA/2)F1 mice (BDF1), which results in acute GVHD, reduced the peripheral blood cell counts, the number of BM cells, and colony-forming unit-granulocyte macrophage (CFU-GM), whereas the injection of 10(8) of DBA/2 cells into BDF1, which results in chronic GVHD, did not affect hematopoiesis 2 weeks after the transfer. To clarify the mechanism of such myelosuppression, we examined the Fas expression in both hematopoietic progenitor cells as well as whole BM cells. The Fas expressions in each fraction significantly increased in BDF1 mice 2 weeks after the induction of acute GVHD, whereas no such effects were observed in the BDF1 mice with chronic GVHD. Furthermore, when such BM cells were incubated with anti-Fas antibody (Jo2), which induces apoptosis through Fas, the fraction of apoptotic cells increased and the number of CFU-GM decreased significantly. The in vivo **administration** of neutralizing anti-**FasL** antibody into BDF1 mice receiving with B6 spleen cells thus protected the host mice from BM failure. These results indicate that the functional expression of Fas on hematopoietic cells plays an essential role in the myelosuppressive effect of GVHD.

L23 ANSWER 7 OF 7 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1997-235546 [21] WPIDS
DOC. NO. NON-CPI: N1997-194816
DOC. NO. CPI: C1997-075485
TITLE: Methods to detect and **treat** **diseases** with **Fas ligand** activity deficiency - by **administering** anti-Fas antibody or **Fas ligand** to increase Fas activity past diseased levels and increasing apoptosis in Fas+ cells.
DERWENT CLASS: B04 S03
INVENTOR(S): KAPLAN, D R
PATENT ASSIGNEE(S): (TKBA-N) TKB ASSOC LP

COUNTRY COUNT: 19
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9712632	A1	19970410	(199721)*	EN	55

RW: AT BE CH DE DK ES FI FR GB GR IE IT LU MC NL PT SE
W: CA JP

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9712632	A1	WO 1996-US15917	19961004

PRIORITY APPLN. INFO: US 1995-4985 19951005
AN 1997-235546 [21] WPIDS
AB WO 9712632 A UPAB: 19970522
Method for treating patients with a deficiency of **Fas ligand** activity comprises the **administration** of an agent to increase **Fas ligand** activity.

Also claimed are:

(1) a method for treating viral infections comprising administering an agent which causes apoptosis of Fas+ cells;

(2) a method for determining when to initiate Fas ligand replacement therapy in a patient by:

(a) determining the Fas ligand activity in the patient, or

(b) determining the presence or amount of Fas ligand in the patient, or

(c) determining the presence or amount of Fas antigen in the patient, and

(3) a method for monitoring the Fas replacement therapy in a patient comprising the processes (a)-(c) as in (2).

USE - The method halts the Fas antigen mediated killing and is therefore useful in diseases such as HIV infection. The Fas ligand activity is increased and allows increased apoptosis to occur. It can also be used to treat Herpes Simplex virus type 2 infections (all claimed), Human herpes virus 6, Epstein-Barr virus and cytomegalovirus. It is also used to bring about natural cell death.

ADVANTAGE - The method increases levels of apoptosis of T-cells using Fas ligand or agonists of the Fas antigens slowing the progress of **disease**. This method minimises toxic effects and cells can be removed, **treated** and re-administered to a patient, where necessary.

Dwg.5a,b/22

FILE 'HOME' ENTERED AT 10:48:07 ON 26 APR 2001